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AN OPTICAL HIGH PRESSURE CELL FOR LIQUID CRYSTALS

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ABSTRACT

A new type of optical high pressure cell for visual observation of phase transitions in liquid crystals under pressure is described. This cell has been useful in identifying the pressure induced mesophases of *p*-methoxy and *p*-ethoxy benzoic acids.

INTRODUCTION

IT was predicted theoretically[1] that certain compounds which do not form liquid crystals at atmospheric pressure can show mesomorphism on application of pressure. Using a differential thermal analysis (DTA) high pressure cell designed by us[2], we have been able to establish experimentally that *p*-methoxy and *p*-ethoxy benzoic acids, which are normally non-mesomorphic, show liquid crystalline behaviour when subjected to pressure[1,3]. Though DTA could unambiguously detect the phase transitions, it was not possible to identify the pressure induced phases. This requires an optical cell wherein the sample may be subjected to pressure and at the same time examined through a polarizing microscope. We have designed and constructed such a cell and taken photographs of the textures exhibited by the pressure induced phases.

DESCRIPTION OF THE CELL

A schematic diagram of the cell is given in Fig. 1. It consists of a stainless steel stationary plate (SP) with a central hole and a coaxial step. A fused silica anvil (A_1) is press fitted into this step. A guard ring (R) also made of stainless steel is shrunk fitted to this anvil by heating both of them to nearly 400° C. This ring has a slightly larger inner diameter towards its top portion so that the other anvil (A_2), also made of fused silica, can just rotate inside the ring. This anvil is fixed in the top stainless steel plate (TP). The two anvils can be pressed against each other by screwing the top plate. A major difficulty that we encountered was to contain the liquid crystal under pressure. After trying out various types of binding rings it was found best to use a teflon spacer (thickness ~ 100 μ) of the type shown in the figure. A special feature of this spacer is that it is machined on both sides to have cone angles which perfectly match the corresponding angles of the anvils. Only then was it possible to contain the sample without leaks.

The cell was enclosed in a small electrical oven provided with windows. The temperature of the sample was probed by a chromel-alumel thermocouple. The entire unit could be placed on the optical stage of a Leitz polarizing microscope equipped with a camera so that observations could be made in transmitted light. As the thickness of the cell was rather large (~ 1″) a low power objective had to be used and consequently this was the limiting factor for the magnification.

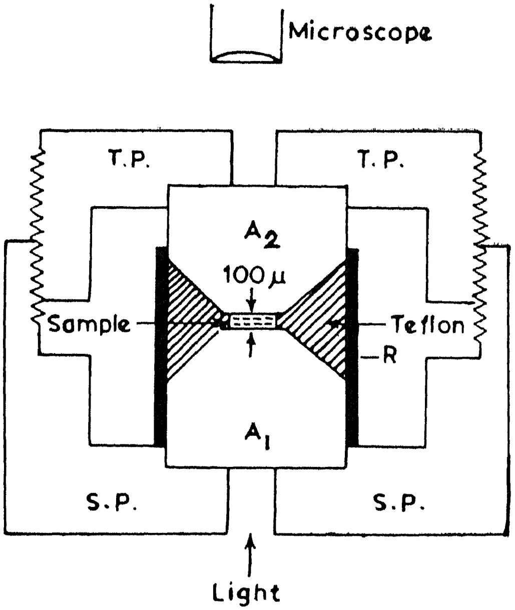

FIG. 1. Schematic diagram of the optical high pressure cell.

IDENTIFICATION OF PRESSURE INDUCED PHASES USING THE CELL

The pressure developed was very roughly estimated in terms of the number of screw turns of the top plate by measuring (by visual observation) the solid-nematic and nematic-isotropic transition temperatures for *p*-azoxyanisole for which dT/dP values are known[1,4,5]. Experiments were then undertaken on *p*-methoxy and *p*-ethoxy benzoic acids to identify the pressure induced phases. The sample which is in the solid phase at room temperature is packed within the spacer. The anvils are gradually pressed together by rotating the top plate. Initially the teflon is compressed but on further turning the sample itself experiences the pressure. Keeping the pressure constant, the temperature is then increased so that the phase transitions are studied at that pressure.

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At low pressures *p*-ethoxy benzoic acid (which at atmospheric pressure has just the solid-isotropic liquid or melting transition at ~ 194° C) shows a single intermediate phase which exhibits the *schlieren* texture typical of a nematic liquid crystal (Fig. 2). At higher pressures it was found that on cooling the isotropic liquid there appears the nematic phase (with a schlieren texture) which at a lower temperature transforms to another phase with 'focal conics' and 'batonnets' characteristic of a smectic liquid crystal (Fig. 3). On further cooling the sample goes over to the solid phase and becomes opaque. It may be mentioned that it was not possible to observe such well defined textures during every experiment. Special care had to be taken to clean the anvil surfaces and to have a uniformly thick sample (~ 100 μ) in order to get good textures.

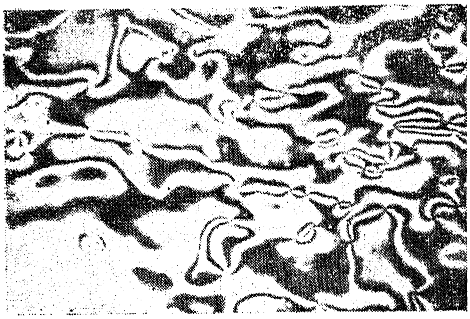

FIG. 2. Schlieren texture exhibited by *p*-ethoxy benzoic acid (crossed polars, × 90).

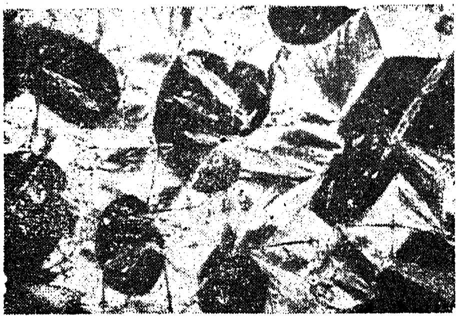

FIG. 3. Focal conics and batonnets shown by *p*-ethoxy benzoic acid (crossed polars, × 90).

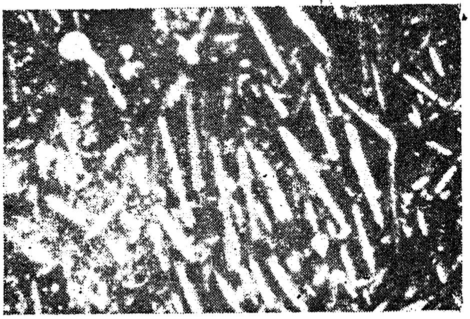

FIG. 4. Batonnets shown by *p*-methoxy benzoic acid (crossed polars, × 90).

p-Methoxy benzoic acid also shows similar textures under pressure. Batonnets shown by the smectic phase of this compound are shown in Fig. 4.

In its present form this cell works for relatively low pressures (of the order of a few hundred bars) only. This is primarily because of the relatively large surface area of the anvil faces which are pressed together, though fused silica itself is capable of withstanding pressures up to 5 kbar. Furthermore, the calibration of pressure is not accurate. Experiments are in progress to modify the design of the cell so as to go to higher pressures and also to improve the accuracy of the pressure calibration.

ACKNOWLEDGEMENTS

We wish to acknowledge the help given by Mr. N. Narayana Iyer and Mr. Chaterjee in the construction of the cell.

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1. Chandrasekhar, S., Ramaseshan, S., Reshamwala, A. S., Sadashiva, B. K., Shashidhar, R. and Surendranath, V., "Proceedings of the International Liquid Crystals Conference, Bangalore, December 3–8, 1973," *Pramāṇa Supplement*, 1, 117.
2. Reshamwala, A. S. and Shashidhar, R. (to be published).
3. Shashidhar, R. and Chandrasekhar, S., *J. de Physique*, 1975, 36, C 1–49.
4. Hulett, G. A., *Z. Phys. Chem.*, 1899, 28, 629.
5. Robberecht, J., *Bull. Soc. Chim. Belge.*, 1936, 37, 597.

DIIODOSALICYLATES OF THE RARE EARTHS

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ABSTRACT

Rare earth diiodosalicylates of the general formula, $Ln(I_2Sal)_3 \cdot xH_2O$, where $x = 5$ when Ln = La, Ce, Pr, Nd; $x = 4$ when Ln = Sm, Ho, Yb and Y, and $I_2Sal = I_2C_6H_2(OH)COO^-$, have been prepared and characterised by chemical analyses, infrared, conductance and differential thermal analyses. The infrared data show that the bonding of the carboxylate group to the metal is bidentate. The infrared and thermal studies reveal that one molecule of water is coordinated to the metal in all the chelates.

INTRODUCTION

THE compounds of salicylic and substituted salicylic acids with metals are of interest because the salicylate ion can attach itself to the metal in a number of ways. A number of salicylates of the rare earths has been synthesised in the past[1,2]. Although studies on a few compounds of rare earths with substituted salicylic acids have been carried out[3,4], no studies have been reported on the preparation and characterisation of the rare earth compounds of 3, 5-diiodosalicylic acid. A few salts of diiodosalicylic acid that have been prepared have been shown to be of considerable biological importance. Bismuth diiodosalicylate has been used in antisyphilitic therapy[5]. The sodium salt has been shown to possess antitoxic properties[6], and has been used for protection from hyperthyreosis[7]. The copper complexes possess pronounced antimicrobial properties[8]. The visible and ultraviolet spectra of copper complexes[9], as also the effect of the lithium salt on the melting points of gelatins[10] have been studied. The present paper deals with the preparation, infrared, conductance and DTA studies on the rare earth diiodosalicylates.

EXPERIMENTAL

Materials.—Diiodosalicylic acid was recrystallised from ethanol before use. Rare earth oxides, 99·9% purity (American Potash and Chemical Corporation, U.S.A.), were treated with aqueous hydrochloric acid and evaporated to dryness to obtain the hydrated rare earth chlorides. Hydrated $CeCl_3$ was prepared similarly by using cerium hydroxide.

Preparation of the Complexes.—Diiodosalicylic acid (2·5 g) was dissolved in ethanol and ethanolic ammonia added drop by drop with stirring, when the ammonium salt of the acid separated out. To a warm aqueous solution of the ammonium salt was added with stirring, an aqueous solution of the rare earth chloride (0·3 g oxide). The thick flocculent precipitate which separated out was filtered, washed with water, dried first in air and then over calcium chloride under reduced pressure.

Analyses.—The metal content of the compounds was estimated by igniting a known weight of the compound to the metal oxide. Carbon and hydrogen in a few complexes were estimated by microanalytical methods by way of verification.

Physical Methods.—The infrared spectra, in Nujol mull, were recorded with a Carl-Zeiss UR-10 automatic spectrophotometer. The molar conductance in acetone was determined by a Siemens conductivity bridge, and the concentration of the solutions was *ca.* 0·001 M. The differential thermograms were recorded on an automatic instrument fitted with chromel-alumel thermocouples, and using calcined alumina as the reference material. The conductance and the infrared data are presented in Table I and Table II respectively.

RESULTS AND DISCUSSION

Analytical results of the compounds conform to the general formula, $Ln(I_2Sal)_3 . xH_2O$, where $x = 5$ when Ln = La, Ce, Pr, Nd and $x = 4$ when Ln = Sm, Ho, Yb and Y. The compounds are quite stable and are unaffected in air. They are insoluble in water, dimethyl formamide, dimethyl sulphoxide and non-polar solvents, but are slightly soluble in ethanol, methanol and acetone.

Unlike the sodium and ammonium diiodosalicylates which are soluble in water, all the rare earth metals form insoluble diiodosalicylates. This may be due to an appreciable covalent character of the metal-carboxylate bond, water being unable to sever this bond. The covalency of the metal-carboxylate bond in these compounds is further confirmed by their conductance measurements in acetone which show their non-electrolyte nature in this solvent.

TABLE I

Analytical and conductance data

Compound	% Metal Found	% Metal Calc.	% Carbon Found	% Carbon Calc.	% Hydrogen Found	% Hydrogen Calc.	Molar Conductance λ, mho cm^2 mol^{-1}
La $(I_2Sal)_3 \cdot 5H_2O$	9·95	9·96	18·20	18·05	1·58	1·36	8·2
Ce $(I_2Sal)_3 \cdot 5H_2O$	9·93	10·02	..	..	..	..	15·4
Pr $(I_2Sal)_3 \cdot 5H_2O$	9·98	10·08	18 14	18·02	1·24	1·36	7·8
Nd $(I_2Sal)_3 \cdot 5H_2O$	10·18	10·30	..	..	..	..	13·9
Sm $(I_2Sal)_3 \cdot 4H_2O$	10·90	10·81	18·32	18·13	1·45	1·22	13·9
Ho $(I_2Sal)_3 \cdot 4H_2O$	11·80	11·76	..	..	..	..	8·2
Yb $(I_2Sal)_3 \cdot 4H_2O$	12·23	12·26	17·92	17·84	1·34	1·20	11·2
Y $(I_2Sal)_3 \cdot 4H_2O$	6·64	6·70	19·30	18·98	1·56	1·28	9·6

Note.- Carbon and hydrogen have been estimated only for a few representative compounds.

TABLE II

Infrared spectral data

I_2SalH	La$(I_2Sal)_3 \cdot 5H_2O$	Na-Salt	Assignments
3520			O-H stretch
	3480		H_2O stretch
	3200		H_2O stretch
1706			C=O stretch
1625	1620	1638	ring C-C stretch
1580			
	1558	1575	asym. COO^- stretch
1445	1450	1450	ring C-C stretch
1420	1430	1430	
1400	1420		
	1370	1368	sym. COO^- stretch
1305			C-O stretch (of COOH)
1240			
1230	1250	1270	C-O stretch (phenolic O)
885			O-H out-of-plane bending (of COOH)
	882	881	C-H out-of-plane bending

Note.—The i.r. spectra of the other diiodosalicylates are quite similar.

The i.r. spectrum of diiodosalicylic acid has not been so far reported in literature. Hence, the frequency assignments for the ligand and the rare earth compounds have been made on the basis of the i.r. data available for benzoic and salicylic acids[11,12]. Of the three possible internally hydrogen bonded forms of salicylic acid, Tsuzuki *et al.*[13] have preferred the one where the phenolic group forms a hydrogen bond with the carbonyl oxygen atom of the cis-carbonyl structure, on the basis of the observation of the bands for carbonyl, carboxylic –OH and phenolic –OH groups at 1690 cm^{-1}, 3530 cm^{-1} and 3200 cm^{-1} respectively, in the i.r. spectrum of salicylic acid. In the present investigation, i.r. bands have been observed for diiodosalicylic acid at 1706 cm^{-1} and 3520 cm^{-1} attributed to the carbonyl and carboxyl –OH stretching vibrations, and another somewhat broadened band in the region 3080–3200 cm^{-1} attributed to the phenolic –OH group. Hence, it can be inferred that diiodosalicylic acid also exists in an internally bonded form which is similar to the one present in salicylic acid.

The two bands at 1706 cm^{-1} and 1310 cm^{-1} which are due to ν C=O and ν C—O of the carboxyl group of the ligand are absent in the rare earth compounds. The rare earth compounds show bands at ~ 1560 cm^{-1} and ~ 1370 cm^{-1} which can be attributed, respectively, to the asymmetric ν COO^- and symmetric ν COO^- vibrational frequencies of the carboxylate group. The corresponding bands for the sodium salt are observed at 1575 cm^{-1} and 1368 cm^{-1}, respectively. Thus the difference between the two ν COO^- frequencies ($\Delta \nu$) is less in the rare earth compounds than in the sodium salt. This shows that, in the compounds investigated, the bonding of the carboxylate group to the metal is not monodentate; if it were so the frequency separation, $\Delta \nu$, would be more than that in the sodium salt. In the case of a bidentate mode of coordination there are two factors influencing the symmetric and asymmetric carboxylate frequencies. Due to the bidentate mode of coordination of the carboxylate group to the metal, there is a decrease of both the COO^- stretching frequencies, as compared to the corresponding value for the sodium salt, due to the drainage of the electron density from the carboxylate group to the metal. But at the same time the O–C–O angle is decreased due to an increase in the metal-oxygen bond strength. This decrease in the O–C–O angle results in a decrease in the frequency separation, $\Delta \nu$[14-16]. These two factors have a unidirectional effect on the asymmetric νCOO^- frequency and an opposite effect on the symmetric νCOO^- frequency. Hence, when the bonding of the carboxylate group is bidentate, the asymmetric frequency decreases and the symmetric frequency shows only small shifts, thus making, $\Delta \nu$, less than that in the sodium salt. Thus the bonding of the carboxylate group, in the compounds investigated, must be bidentate, as $\Delta \nu$ has been found to be less in these compounds than in the sodium salt. However, in the present chelates, the shift of the asymmetric frequency is less than the corresponding shift in the rare earth salicylates[2], suggesting that the metal-carboxylate bond has more ionic character in the former than in the latter.

The –COOH group gives bands at 3520 cm^{-1} (s) and 2600 cm^{-1} (w) due to the –OH stretching vibrations and another broad band at 885 cm^{-1} due to the –OH out-of-plane vibration. These bands are absent in the chelates.

The rare earth chelates show bands in the 3000–3600 cm^{-1} region due to the stretching modes of water molecules. The bands in this region can be assigned to the lattice water ($\sim$ 3475 cm^{-1}) and to the coordinated water ($\sim$ 3,200 cm^{-1}).

Differential thermograms were obtained for La, Nd and Sm diiodosalicylates. They show similar behaviour for the three compounds studied and the same behaviour can be expected for the other rare earth compounds as well. An endotherm in the region 70–100° C shows the removal of water. Heating of the samples at 100° C for 4–5 hours showed that four water molecules were removed in the case of La, Ce, Pr and Nd chelates, and three water molecules in the case of the Sm, Ho, Yb and Y chelates. Thus one molecule of water is coordinated in all the chelates, the rest of them being only lattice held. There is no endotherm in the differential thermograms showing the removal of the coordinated water. The removal of the coordinated water is probably accompanied by the decomposition of the compound which gives a strong and broad exotherm centered around 300° C.

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1. Misra, S. N., *Indian J. Chem.*, 1967, **5**, 508.
2. Kanekar, C. R., Marathe, V. R. and Thakur, N. V., *Proc. Chem. Symp., DAE India*, 1969, 2, 265.
3. Spacu, P., Antonescu, E., Mavrodin, M. and Serban, S., *Acad. Rep. Populare Romine, Studii. Cercetari. Chim.*, 1963, **11**, 385.
4. Shestakova, M. T. and Pirkes, S. B., *Zh. Neorg. Khim.*, 1970, **15**, 993.
5. Pollano, F., *Minerva med.*, 1930, **1**, 786.
6. Velluz, L., *Compt. rend.*, 1931, **193**, 372.
7. Marsal, A., *Rev. Soc. Argentina Biol.*, 1936, **12**, 151.
8. Gershan, H., Parmegiani, R. and Nickerson, W. J., *Appl. Microbiol.*, 1962, **10**, 556.
9. Schulman, S. G., Kilroy, W. P. and Gershan, H., *J. Phys. Chem.*, 1968, **72**, 3372.
10. Bello, J., Riese, H. C. A. and Vinograd, J. R., *Ibid.*, 1956, **60**, 1299.
11. Taylor, M. D., Carter, C. P. and Wynter, C. I., *J. Inorg. Nucl. Chem.*, 1968, **30**, 1503.
12. Green, J. H. S., Kynaston, W. and Lindsey, A. S., *Spectrochim. Acta*, 1961, **17**, 486.
13. Mori, N., Asano, Y., Irie, T. and Tsuzuki, Y., *Bull. Chem. Soc. Jap.*, 1969, **42**, 484.
14. Spinner, E., *J. Chem. Soc.*, 1964, p. 4217.
15. Grigor'ev, A. I., *Russ. J. Inorg. Chem.*, 1963, **8**, 407.
16. —, and Maksimov, V. N., *Ibid.*, 1964, **9**, 580.

CADMIUM (II) COMPLEXES WITH AROYL HYDRAZONES

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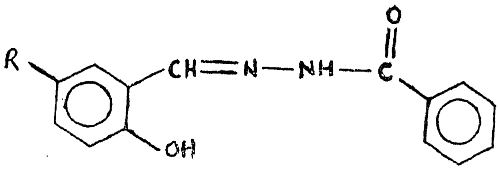
ABSTRACT

Eight complexes of Cd(II) have been prepared by reacting cadmium acetate with aroyl hydrazones in aqueous ethanolic medium. The elemental analyses indicate 1 : 2 stoichiometry. On the basis of infrared spectral studies, it has been shown that the ligands react in the *keto form* and the complexes have coordination number six.

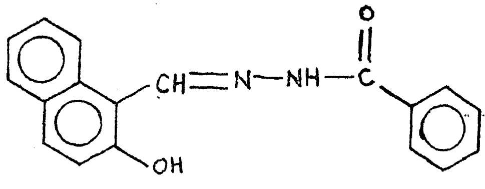
INTRODUCTION

THE hydrazides have been successfully tried for the complexation and they have been shown to coordinate through C=O and the terminal NH_2 groups[1]. The hydrazones, containing C=O and C=N coordinating sites, are the reaction products of hydrazides and aldehydes or ketones[2]. The *o*-hydroxy aldehydes or ketones yield the bases containing active –OH group in addition to the C=O and C=N coordinating groups. The earlier literature[1,3] displays that these bases show monovalent bidentate behaviour. However, a few complexes with salicylhydrazone and isopropyl salicylhydrazone are reported in the literature, in which –OH group remains inert to the complex formation[4]. All this information stands as a testimony for the versatility of the hydrazones as chelating agents. However, the work on the complexes of divalent metal ions with hydrazones is limited.

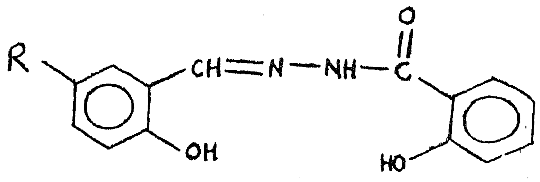
The present work embodies the synthesis and spectral properties of Cd(II) complexes with the following hydrazones :



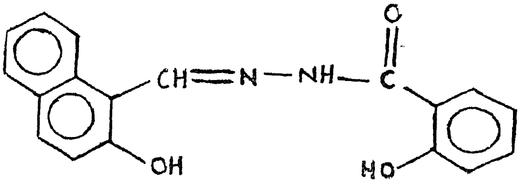
I R=H
II R=CH_3
III R=Cl



IV



V R=H
VI R=CH_3
VII R=Cl



VIII

EXPERIMENTAL

Materials and Method

Ethylbenzoate, methylsalicylate and hydrazine hydrate used for preparing the hydrazides were of reagent grade. Cadmium acetate was an analytical reagent. Benzoyl hydrazide and *o*-hydroxy-benzoylhydrazide were prepared according to standard methods[5]. The substituted aldehydes were prepared according to Duff's method[6]. The hydrazones were prepared by the reaction of benzoylhydrazide or *o*-hydroxy-benzoylhydrazide (0·1 M) with corresponding aldehyde (0·1 M) in ethanol.

Preparation of Complexes

To an ethanolic solution containing (0·2 M) of hydrazide and aldehyde, an aqueous cadmium acetate solution (0·1 M) was added. The reaction mixture was heated for a while on water bath and the complex precipitated was filtered and washed free from the ligand with ethanol and finally with ether and dried in vacuum over P_2O_5.

Elemental Analysis

Cadmium in the complexes was estimated gravimetrically as cadmium pyrophosphate[7] and nitrogen was estimated by Dumas method. The results of the elemental analysis are shown in Table I.

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* For correspondence.

TABLE I

Elemental analysis of Cd(II) complexes with aroyl hydrazones

Sl. No.	Ligand No.	Complex No.	Empirical formula	% Cd		% N	
				Found	Calcd.	Found	Calcd.
1.	I	IX	$(C_{14}H_{11}N_2O_2)_2$ Cd	19·77	19·03	10·11	9·48
2.	II	X	$(C_{15}H_{13}N_2O_2)_2$ Cd	17·50	18·18	9·10	9·06
3.	III	XI	$(C_{14}H_{10}N_2O_2Cl)_2$ Cd	16·79	17·04	8·95	8·49
4.	IV	XII	$(C_{18}H_{13}N_2O_2)_2$ Cd	16·05	16·29	8·54	8·11
5.	V	XIII	$(C_{14}H_{11}N_2O_3)_2$ Cd	17·45	18·05	9·11	9·00
6.	VI	XIV	$(C_{15}H_{13}N_2O_3)_2$ Cd	17·66	17·28	8·92	8·61
7.	VII	XV	$(C_{14}H_{10}N_2O_3Cl)_2$ Cd	16·80	16·26	7·89	8·10
8.	VIII	XVI	$(C_{18}H_{13}N_2O_3)_2$ Cd	16·10	15·56	7·35	7·75

Physical Measurements

The conductance measurements were done on an ELICO CM–82 conductivity bridge with a cell having cell constant 0·829 cm^{-1}. The IR spectra of the ligands and the complexes in nujol mull were recorded on Carlzeiss UR–10 recording spectrophotometer in the region of 4000–400 cm^{-1}.

RESULTS AND DISCUSSION

The complexes are yellow in colour, insoluble in common organic solvents. To some extent they are soluble in DMF and DMSO. The results of the elemental analysis (Table I) show that the complexes have 1 : 2 stoichiometry.

Conductance

The molar conductance in DMSO at the concentration $\sim 10^{-3}$ M is too small to measure. Hence the complexes are non-electrolytes in this solvent.

Infrared Spectra

The important infrared frequencies of the complexes are shown in Table II.

All the complexes show medium intensity broad band in the region 3225–3150 cm^{-1} due to NH stretch[8]. The complexes formed of the ligands I–IV, do not display any band in the region 2800–2700 cm^{-1}, whereas, the complexes of the ligands V–VIII have broad weak band around 2700 cm^{-1} characteristic of intramolecular H-bonded OH[9]. The bands of the ligands associated with the C=O and C=N stretching vibrations (Table II) show measurable shift towards low frequency in the complexes supporting the view that both C=O and C=N are involved in the coordinate bond formation. The three high intensity bands in the region 1600–1500 cm^{-1} are due to the aromatic C=C stretching vibrations.

All the complexes show high frequency shift of the band around 1280 cm^{-1} attributable to the phenolic C—O stretch. In complexes XIII to XVI, there is a band around 1280 cm^{-1} having strong intensity. This is assigned to the phenolic C—O due to salicyloylhydrazide moiety.

These observations make us to envisage that the ligands I–VIII exist in the *keto form* in the complexes.

Of the several bands observed in the 600–400 cm^{-1} region which appear unchanged in the intensity and positions are due to the ligand skeletal vibrations. It is rather difficult to make the conclusive assignments amidst the high intensity skeletal bands which interfere with the M—N and M—O and M←O vibrations. Hence the assignments made here are based on the reported data and are tentative.

Amongst the three intense bands which appear in the 600–500 cm^{-1} region the band around 535 cm^{-1} is assigned to the Cd–N stretch as it shows little variation whereas the remaining bands are inert to the complexation. This assignment closely agrees with the previous assignments[10,11]. The band observed in the region 490–460 cm^{-1} is an intense one and is assigned to the Cd←O band in view of the previous assignments[1]. The band observed in the region 445–430 cm^{-1}, on analogy with the previous data[11,12], is assigned to the Cd—O stretch. All these bands show little variation in their positions indicating that all these complexes have the same coordination number.

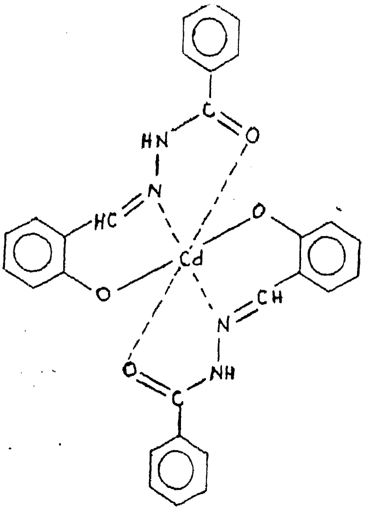
On the basis of analytical data, infrared spectral results and the known hexacovalency of cadmium (II), the following structure may be proposed.

TABLE II

Important infrared frequencies (in cm^{-1}) of Cd (II) complexes with aroyl hydrazones and their assignments

Sl. No.	Ligand No.	Complex No.	ν(NH)	ν(C=O)	ν(C=N)	ν(C=C)	ν(C–O)	ν(Cd–N)	ν(Cd←O) / ν(Cd—O)
1.	I	IX	3150 m	1625 s	1575 s	1575 s 1555 s 1500 m	1305 s	530 s	460 s 430 s
2.	II	X	3160 m	1630 s	1575 s	1575 s 1555 s 1500 w	1325 s	540 s	460 s 430 s
3.	III	XI	3150 m	1625 s	1580 s	1580 s 1545 s 1500 m	1325 s	535 s	465 s 430 s
4.	IV	XII	3160 m	1625 s	1585 s	1575 s 1560 s 1500 s	1325 s	530 s	470 s 445 s
5.	V	XIII	3220 m	1625 s	1580 s	1580 s 1565 s 1500 w	1340 s 1280 m	535 s	480 s 445 s
6.	VI	XIV	3150 m	1620 s	1575 s	1575 s 1555 w 1480 s	1280 s(br)	535 s	490 s 445 s
7.	VII	XV	3150 m	1625 s	1575 s	1575 s 1500 w	1335 s 1290 m	530 s	460 s —
8.	VIII	XVI	3225 m	1625 s	1585 s	1585 s 1565 s 1510 w	1320 m 1300 s	530 s	475 s 430 s

In the ligands the following assignments may be made: ν(NH) ~ 3200 cm^{-1}, ν(intramolecular H-bonded OH) 2800–2700 cm^{-1}, ν(C=O) 1670–1650 cm^{-1}, ν(C=N) 1625–1610 cm^{-1} and ν(C—O) ~ 1280 cm^{-1}.



ACKNOWLEDGEMENTS

One of the authors (B. R. H.) is thankful to Prof. E. S. Jayadevappa, Head of the Department of Chemistry, Karnatak University, Dharwar, for laboratory facilities and encouragement. Thanks are also due to Dr. V. H. Kulkarni for his help.

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1. Narang, K. K. and Miss Aggarwal, A., *Inorg. Chim. Acta*, 1974, 9, 137.
2. Ohta, H., *Bull. Chem. Soc.* (*Jap.*), 1958, 31, 1056; *Ibid.*, 1960, 33, 2.
3. El-Sayed, L. and Iskander, M. F., *J. Inorg. Nucl. Chem.*, 1971, 33, 435.
4. Issa, R. M., Iskander, M. F. and El-Shazly, M. F., *Z. anorg. allg. Chem.*, 1967, 354, 98.
5. Vogel, A. I., *A Text Book of Practical Organic Chemistry*, Longmans, Green, London, 1968.
6. Duff, J. C., *J. Chem. Soc.*, 1941, p. 547.
7. Vogel, A. I., *A Text Book of Quantitative Inorganic Analysis*, Longmans, Green, London, 1969.
8. Rao, C. N. R., *Chemical Applications of Infrared Spectroscopy*, Academic Press, New York, London, 1962.
9. Biradar, N. S. and Kulkarni, V. H., *J. Inorg. Nucl. Chem.*, 1971, 33, 2451.
10. Nakamoto, K., *Advances in the Chemistry of Coordination Compounds*, Ed. Kirschner, S., Macmillan, New York, 1961.
11. Biradar, N. S., Mahale, V. B. and Kulkarni, V. H., *Inorg. Chim. Acta*, 1973, 7, 267; *Inorg. Nucl. Chem. Lett.*, 1972, 8, 997.
12. Nakamoto, K., McCarthy, P. J. and Martell, A. E., *J. Am. Chem. Soc.*, 1961, 83, 1272.

DEEP ELECTRICAL RESISTIVITY SOUNDINGS IN THE DECCAN TRAP REGION

L. N. KAILASAM, A. G. B. REDDI, M. V. JOGA RAO, K. SATHYAMURTHY AND B. S. R. MURTY
Geological Survey of India

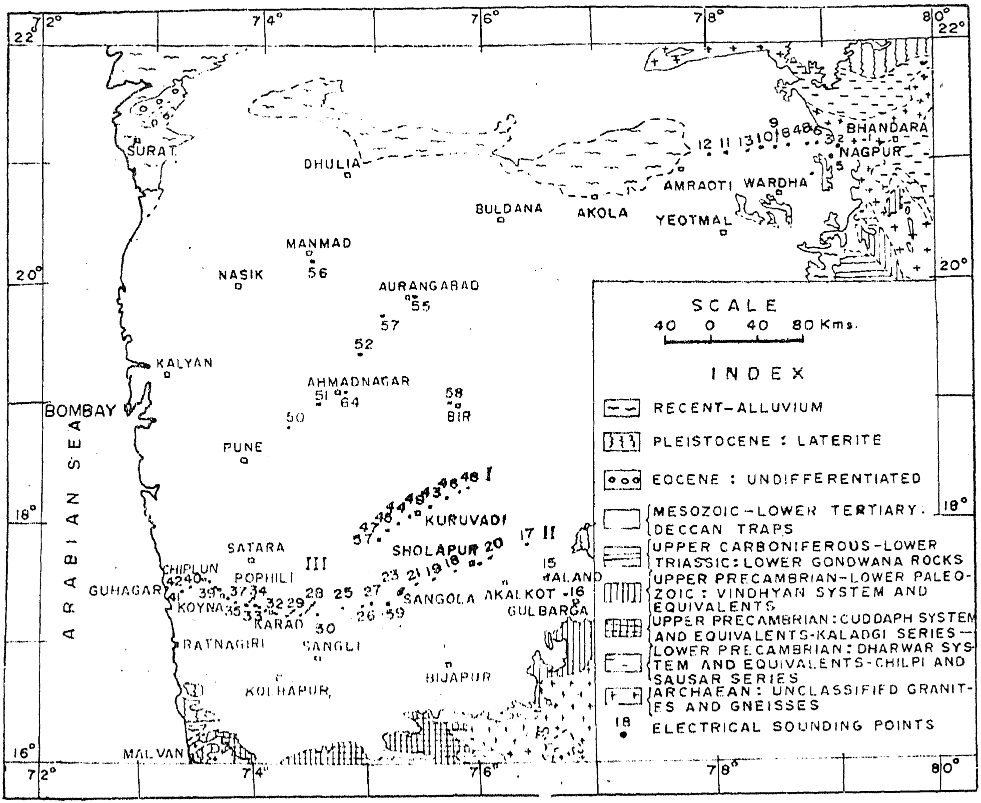
REGIONAL gravity surveys of the Deccan trap region in western and central India were conducted by the Geological Survey of India over a period of eight years (1964–72) in connection with the International Upper Mantle Project and more than 400,000 square kilometres have been covered so far by these surveys which are now being continued in the areas to the north of the Purna, Tapti and Narmada valleys. These investigations had thus been initiated quite some time prior to the devastating Koyna earthquake of 1967 which shook some of the complacent ideas regarding the stability of the Indian peninsular shield. The gravity surveys were supplemented by spot seismic soundings at a number of selected points over the entire region covered by the gravity observations. Detailed gravity-*cum*-refraction seismic surveys were also conducted in the Koyna region to unravel the deep subsurface tectonic features having a possible bearing on the prevailing seismicity in this region. The results of these investigations have been presented in a number of published papers (Kailasam *et al.*, 1969, 1972). These surveys, besides bringing out a number of interesting and significant gravity features of deep seated causes and of tectonic significance, have also enabled the estimation, by refraction seismic soundings, of the overall thickness of the Deccan trap at a number of points.

The Bouguer and residual gravity maps have brought out a number of pronounced gravity features. The Bouguer gravity map (Kailasam *et al.*, 1972) presents some prominent gravity 'highs' in the Sangola and Nasik regions and marked zones of gravity 'low' in the Koyna–Karad and Kurudwadi–Dhond regions, as also another zone of pronounced gravity 'low' adjacent to and north of the gravity 'high' near Nasik. These pronounced gravity 'highs' and 'lows' have been interpreted as indicative of zones of marked uplifts and subsidence of a deep-seated, crustal nature.

The refraction seismic soundings have indicated an overall depth to the base of the trap of the order of 100 to 200 metres beneath the ground surface over the southern and eastern margins of the Deccan trap region, covered by these surveys while maximum depths of the order of 1,100 to 1,200 metres have been indicated in the western and northwestern parts of Maharashtra. The seismic results have further indicated a macroscopic longitudinal wave velocity varying generally from 4·8 to 5·2 km/sec. for the Deccan trap, whereas a longitudinal velocity of 5·8 to 6·4. km/sec. has been indicated for the sub-trap horizon. It was, however, observed that, while at most of the seismic sounding points the travel-time curves yielded reliable estimates to the base of the Deccan trap, yet at a few other points some ambiguity arose in the identification of the layer below the velocity interface, presumed to correspond to the base of the trap due to the overlapping seismic velocities of the Archaean and pre-Cambrian basement rocks (including limestones, quartzites, slates etc., belonging to the Kaladgi and Bhima series of pre-Cambrian age). This was further complicated by the substantially higher seismic velocity of well over 6 km/sec. exibited by the trap formation itself at depth in some parts of the area—a fact established by extensive laboratory determinations of the longitudinal wave velocity in a number of Deccan trap samples. Such high velocities were especially observed in Deccan trap samples relatively rich in olivine and these samples also showed a higher density of over 3 g/cc. It was, therefore, decided to supplement the results of the refraction seismic sounding by deep electrical resistivity depth probes in view of the fact that the resistivity contrast between the Deccan trap on the one hand, and the pre-Cambrian gneisses and the Kaladgis on the other is of the order of 1 : 5. The absolute resistivity value of the Deccan trap formation is of a low order of 50 to 200 ohm-meters while the true resistivity of the pre-Cambrian rocks, such as the gneisses and the Kaladgi limestones and quartzites, is well over 1,000 ohm-meters, thus enabling a clear identification of the Deccan trap from the latter formations, although the resistivity results do not enable us to distinguish between pre-Cambrian gneisses and Kaladgi limestones or quartzites as all these pre-Cambrian formations show a high order of overlapping resistivity.

A number of deep electical resistivity soundings were conducted employing a modern, sophisticated type of Deep Resistivity unit (DRS–7) manufactured by Scintrex, Canada, with an effective depth of exploration of more than 2,000 metres. These electrical soundings were carried out at a number of selected points distributed over the trap territory as well as along an east-west traverse of roughly 360 km in length from Sholapur on the east to Guhagar on the west coast passing through the marked gravity 'high' in the Sangola area and the pronounced gravity 'low' in the Karad-Koyna region, as also along two short traverses, one in the Kurudwadi area and the other between Amaraoti and Nagpur (Fig. 1). The sounding profiles were conducted employing the Schlumberger array of current and potential electrodes with maximum current electrode spreads 'AB' of 4,000 m.

zones are of special interest from the point of view of ground water exploration, as these conductive zones are often suggestive of intertrappean horizons or vesicular portions within the trap. Such significant conductive zones indicative of ground water potentialities are brought out, for instance, in the sounding curve near Sholapur at a depth interval of roughly 30 to 200 metres (Fig. 2) and in the resistivity curve for sounding station, 34 to the west of Karad at a depth interval of roughly 50 to 110 metres (Fig. 3) from the ground surface.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18 ELECTRICAL SOUNDING POINTS

FIG. 1

Typical resistivity curves are presented for some of the sounding stations in Figs. 2 and 3, in which the computed depths to the successive resistivity interfaces are shown alongside the abscissa scale of half current electrode spread. One characteristic feature to be observed is that the Deccan trap formation as a whole shows marked resistivity stratification, apparently corresponding to lithological stratification including intertrappean formations and vesicular portions within the trap itself, presenting, however, on overall low resistivity for the Deccan trap section as a whole in relation to the highly resistive sub-trap basement. Such marked vertical variations in resistivity are restricted generally to the top one or two hundred metres of the trap within which, incidentally, conductive

The values of the overall thickness of the Deccan trap deducted from the deep resistivity soundings at some of the stations are presented in Table I together with the seismically indicated depths.

It may be noted that there is generally a fair agreement between the electrically and seismically determined depths, allowing for the margin of discrepancy caused by such factors as stratification and macro-anisotropy (electrical) within the trap involved in the electrical resistivity method which may vitiate the computed depths by a margin of 15% relative to the true depths. Also, the resistivity and seismic sounding stations do not coincide [illegible] that are [illegible] Even making all [illegible] may be noted that [illegible] between the [illegible] depths [illegible] being much [illegible] depth. In [illegible] that the [illegible] trap has [illegible] more than [illegible] of velocity [illegible] ment [illegible] has [illegible] where a [illegible] 200 meters was [illegible] collected at depths from [illegible] tudinal velocity of [illegible] Geophysical Laboratory [illegible]

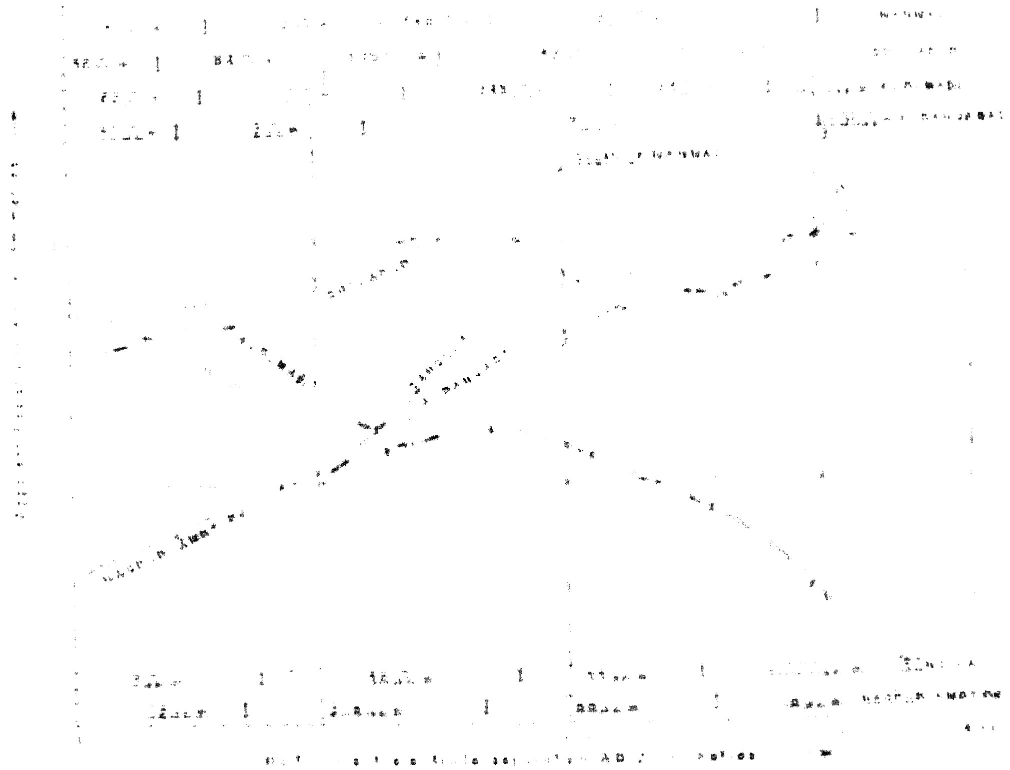
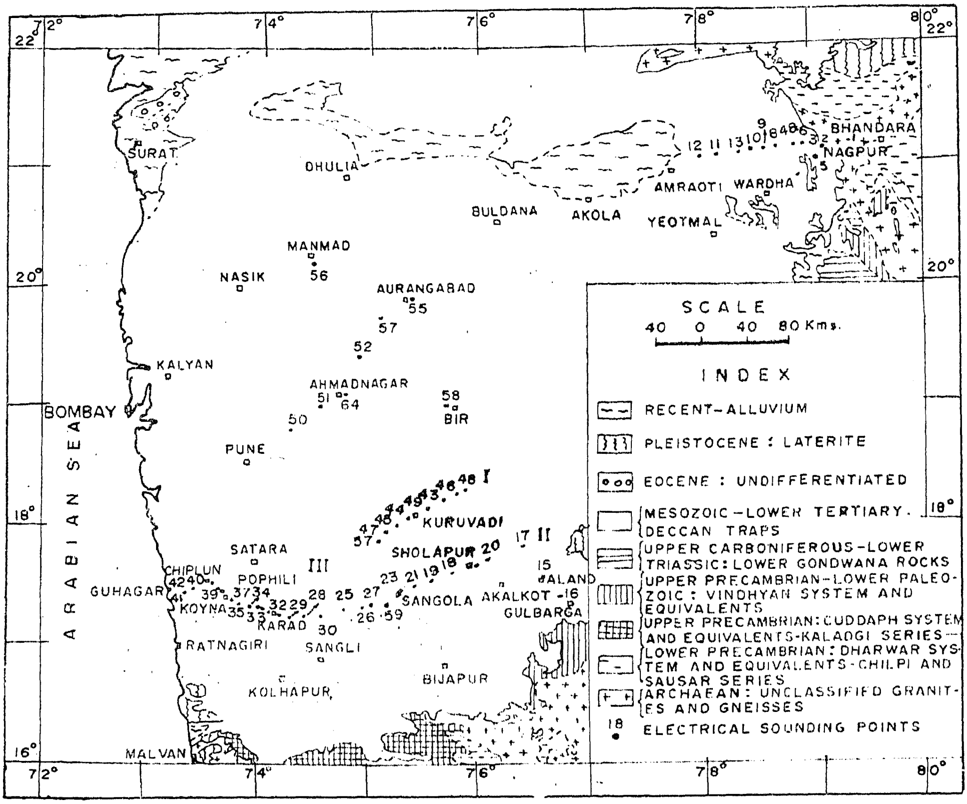


Fig. 2. Typical electrical resistivity soundings [illegible] the Deccan Trap [illegible]

The results of the resistivity soundings along the [illegible] traverse from Sholapur to Gulbarga (Fig. 4) are of particular interest. The base of the Deccan Trap column as a whole is shown in the section along the traverse presented in Fig. 4, which brings out [illegible] as suggested by Krishna Brahmam [illegible] the Karad Koyna [illegible] has been indicated by the results [illegible] data as [illegible] the Koyna [illegible] area. In fact, the [illegible] map presented by Kailasam marked gravity 'high' in the Sangola area and the pronounced gravity 'low' in the Karad-Koyna region, as also along two short traverses, one in the Kurudwadi area and the other between Amaraoti and Nagpur (Fig. 1). The sounding profiles were conducted employing the Schlumberger array of current and potential electrodes with maximum current electrode spreads 'AB' of 4,000 m.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The values of the overall thickness of the Deccan trap deducted from the deep resistivity soundings at some of the stations are presented in Table I together with the seismically indicated depths.

It may be noted that there is generally a fair agreement between the electrically and seismically determined depths, allowing for the margin of discrepancy caused by such factors as stratification and macro-anisotropy (electrical) within the trap involved in the electrical resistivity method which may vitiate the computed depths by a margin of 15% relative to the true depths. Also, the resistivity and seismic sounding stations do not coincide exactly but are shifted by a few hundred metres. Even making an allowance for these factors, it may be noted that at a few stations, the discrepancy between the electrically and seismically computed depths is considerable, the seismically deduced depth being much smaller than the electrically computed depth. In such cases, it would be reasonable to infer that the seismically computed depth corresponds to trap horizon itself, with a higher seismic velocity of more than 6 km/sec. which is of the same order of velocity of the Archaean and Pre-Cambrian basement formations mentioned earlier. In fact, this has been proved to be the case at Alore, some 6 km to the north-northwest of Pophli (Fig. 1) where a drill hole taken to depths of more than 200 metres was still within the traps. Core samples collected at depths from this drill hole showed longitudinal velocity of 6·3 km/sec. as determined in the Geophysics Laboratory of the G.S.I.

some intresting and significant features. In the region of the Sangola gravity 'high', the trap thickness is indicated to be relatively quite small conforming to a crustal uplift in this region as inferred from the gravity anomaly map, whereas in the Karad-Koyna area further to the west, a marked thickness of well over 1,000 metres of trap is indicated beneath the ground surface in the form of a synclinal sag conforming to a zone of subsidence (Kailasam *et al.*, 1972). Another noteworthy feature is that, between Koyna and Pophli to the west of the trap scarp, an abrupt fall in the trap base of roughly 400 metres is indicated, suggestive of a fault postulated earlier on the basis of the gravity and seismic results (Kailasam *et al.*, 1969). Further to the west along the section, between Chiplun and Rampur, another fault is suggested, also as deduced earlier on the basis of the gravity and seismic data. It is also noteworthy that no major rift, as suggeted by Krishna Brahmam *et al.* (1973), corresponding to the Karad-Koyna gravity 'low' has been indicated by the resistivity data, as also in the Kurudwadi area. In fact, the Bouguer gravity map presented by Kailasam

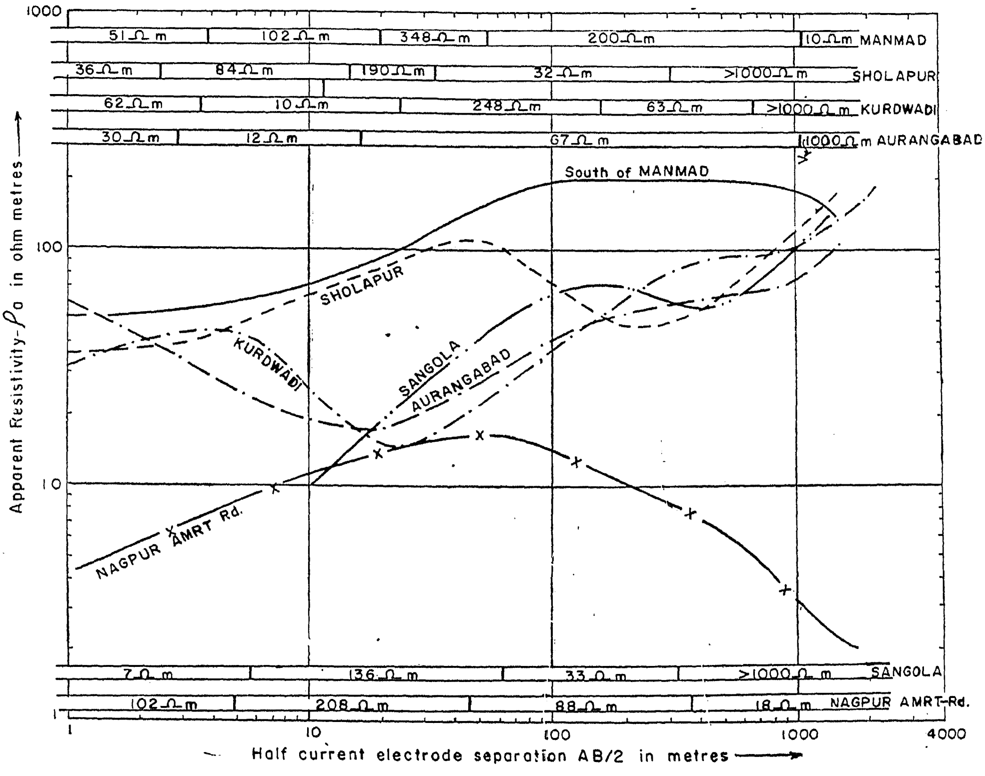

FIG. 2. Typical electrical resistivity soundings with results over the Deccan Trap in general.

The results of the resistivity sounding along the traverse from Sholapur to Guhagar (Fig. 4) are of particular interest. The base of the Deccan trap column as a whole is shown in the section along this traverse presented in Fig. 4, which brings out

TABLE I

Stn. No.	Locality	Depth from ground surface to base of trap/high resistivity layer from resistivity soundings (m)	Depth from ground surface to base of trap/high velocity interface from seismic profiles (m)
60	Sholapur	290	260
55	Aurangabad	990	915
31	Karad	670	500
38	Pophli	530	200
41	West of Rampur	810	670
57	Velapur	580	290
59	Sangola	327	..
44	Kurudwadi	680	..
56	South of Manmad	>1000	..
4	Nagpur–Amraoti road	>1000	..

et al. (1972) and the subsequent detailed gravity surveys conducted by the GSI in this region do not at all indicate such a rift. The gravity gradient to the west of Koyna is quite steep, being roughly 3 milligals per kilometre and at some places even much higher, whereas to the east of Karad, the gravity gradient has the normal small regional magnitude of 0·5 milligal per kilometre. Furthermore, the pronounced gravity 'low' in the Koyna area is in the form of a closed, fairly restricted feature and does not conform to an extended, linear feature of a rift. The resistivity depths to the base of the trap along the Sholapur–Guhagar traverse thus bear out the earlier results and features brought out by the gravity and seismic surveys. Of particular significance is the fact that, in the Koyna–Karad region of pronounced gravity 'low', a thick section of trap is indicated, whereas in the zone of gravity 'high' in the Sangola area, a small thickness of trap is indicated, thereby establishing that the pronounced gravity anomalies are of deep-seated causes due to uplifts and subsidence within the deep crustal and crust-mantle layers. The system of parallel faults and associated fracturing indicated to the west of Koyna has

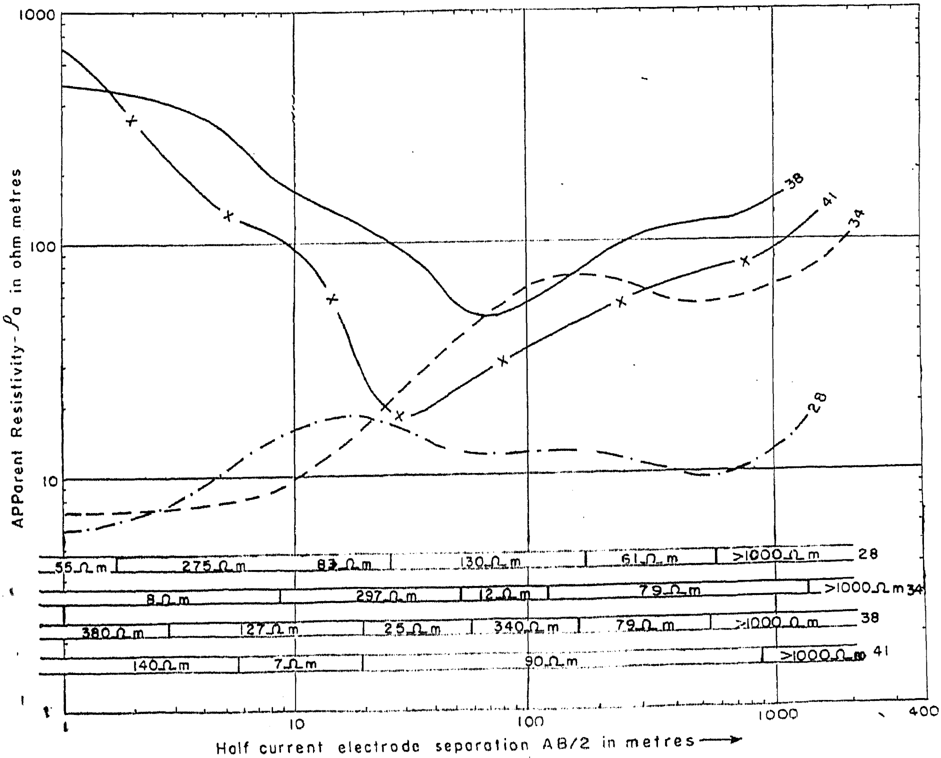

FIG. 3. Typical electrical resistivity soundings along Sholapur–Karad–Guhagar traverse.

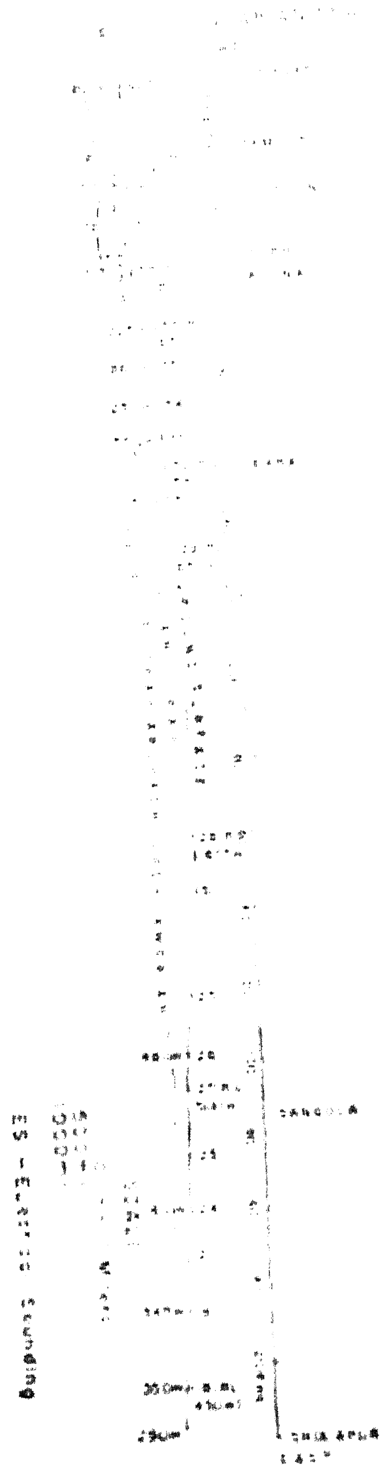

FIG. 4

[illegible] of the Deccan Trap and investigating the sub-trappean [illegible]

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2. [illegible] Upper Mantle, 1969, 100, [illegible]
3. [illegible] Ibid. Upper Mantle, 1969, 100, [illegible]
4. Murthy, B. G. K. and [illegible] A. Y. S. R., [illegible] 1972, 42, [illegible]
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56	South of Manmad	>1000	..
4	Nagpur–Amraoti road	>1000	..

et al. (1972) and the subsequent detailed gravity surveys conducted by the GSI in this region do not at all indicate such a rift. The gravity gradient to the west of Koyna is quite steep, being roughly 3 milligals per kilometre and at some places even much higher, whereas to the east of Karad, the gravity gradient has the normal small regional magnitude of 0·5 milligal per kilometre. Furthermore, the pronounced gravity 'low' in the Koyna area is in the form of a closed, fairly restricted feature and does not conform to an extended, linear feature of a rift. The resistivity depths to the base of the trap along the Sholapur–Guhagar traverse thus bear out the earlier results and features brought out by the gravity and seismic surveys. Of particular significance is the fact that, in the Koyna–Karad region of pronounced gravity 'low', a thick section of trap is indicated, whereas in the zone of gravity 'high' in the Sangola area, a small thickness of trap is indicated, thereby establishing that the pronounced gravity anomalies are of deep-seated causes due to uplifts and subsidence within the deep crustal and crust-mantle layers. The system of parallel faults and associated fracturing indicated to the west of Koyna has

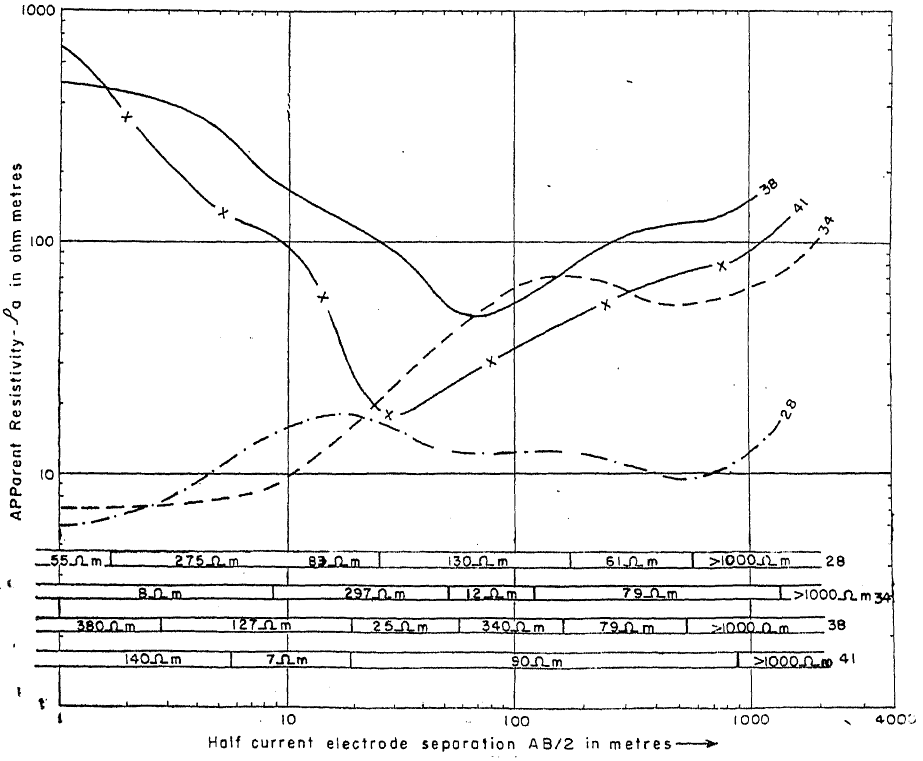

FIG. 3. Typical electrical resistivity soundings along Sholapur–Karad–Guhagar traverse.

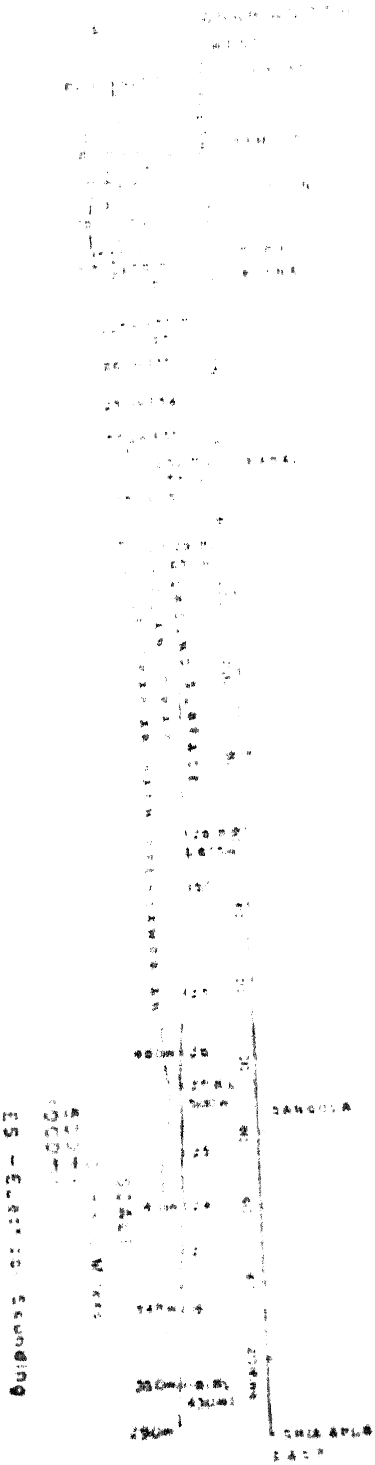

FIG. 4

[illegible] of a deep [illegible] Deccan Trap [illegible] Bhima [illegible] 1970.

[illegible] Deccan Trap and investigating the sub-trappean [illegible].

The authors are thankful [illegible] the publication of this paper.

1. [illegible] Murthy, B. G. K. [illegible] of India [illegible] Upper Mantle, 1970, 100, [illegible].

2. [illegible] S. M. and Sinha, B. B. M. [illegible] Upper Mantle, 1970, 100, [illegible].

3. [illegible] and [illegible] Upper Mantle, 1969, 100, [illegible].

4. Murthy, B. G. K. and [illegible] A. Y. S. R., [illegible] 1972, 42, [illegible].

5. [illegible] Research Bulletin, 1974, [illegible].

FIRST INTERNATIONAL SYMPOSIUM ON INVERTEBRATE REPRODUCTION

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THE vast majority of animals in the world are invertebrates, but paradoxically enough, most studies on reproductive physiology deal with vertebrates. Reproduction of vertebrates is receiving greater attention probably because of the closer affinity of man to vertebrates, and also because most of his domesticated livestock are vertebrates. Invertebrates have an equally strong case inasmuch as they exhibit a wide variety of reproductive patterns and behaviour and there are many species among invertebrates that are useful or harmful to mankind. The International Symposium on Reproductive Physiology of Invertebrates (ISRPI) held at Calicut from September 10 to 12, 1975 under the joint auspices of the University of Calicut and the Kerala Academy of Biology considered the various aspects of the physiology of sex and reproduction in a variety of invertebrate groups. Of the 66 contributions from 14 countries, 38 were presented on the floor. ISRPI was inaugurated in the Library Hall of Calicut University by Prof. Sukumar Azhikode, Acting Vice-Chancellor of the University. N. B. Nair, president of the Kerala Academy of Biology, who presided over the function stressed the importance of studies on reproduction of marine invertebrates.

K. G. Adiyodi, convener of ISRPI in his welcome address, "New perspectives and challenges", pointed out the need for studies on the reproductive physiology of lowly organised invertebrates. These investigations could prove valuable in tracing the physiological phylogeny of reproduction, and also the relation between somatic growth and asexual and sexual types of reproduction. There is great need to look for extraovarian origin of yolk in invertebrates (that oviposit frequently or lay eggs in large numbers) other than insects and crustaceans. The site of synthesis of vitellogenins in crustaceans is also to be elucidated. Studies on the modes of action of invertebrate gonadotropins have suffered as no gonadotropin other than the insect juvenile hormone has been characterized todate. K. G. Adiyodi also suggested that the physiology and control of ovoviviparity and viviparity in invertebrates could make interesting comparisons with those of vertebrates.

There were eight scientific sessions (name of chairman in brackets): Sexual physiology and population dynamics (T. K. Watanabe, Japan), Spermatogenesis (W. H. Clark, Jr., USA), Sperm motility, egg cortical reaction and fertilization (Richard L. Miller, USA), Ovarian development and reproductive cycles (R. Nagabhushanam, India), Reproductive ecology and biochemistry (K. Hotta, Japan), Reproductive tracts and accessory sex glands (D. F. Went, Switzerland), Reproductive behaviour (B. R. Laurence, UK) and Mechanisms controlling reproduction (T. S. Adams, USA). Genetic variations and productivity in natural populations of *Drosophila melanogaster* were discussed by T. K. Watanabe (Japan). Katsunuma population has maintained for many years several polymorphic inversions; the heterokaryotype showed significant superiority with regard to productivity over both homokaryotypes. V. K. Bhasin and M. K. K. Pillai (India) suggested that mosquitoes can be more effectively sterilized by slower and sustained penetration of phosphoramides at low doses; in laboratory experiments, the sexual behaviour of male *Aedes aegypti* sterilized by phosphoramide treatment was not found altered. R. J. Irving-Bell's (Australia) studies on *Culex pipiens* complex indicate that cytoplasmic incompatibility may only occur between wild populations from climatically different environments rather than with geographical distance. Aspects of spermatogenesis studied by electron microscopy were presented by J. Pochon-Masson and I. D. G. Ginneken (France) and Y. Takahashi *et al.* (Japan). Richard L. Miller (USA) who reviewed the distribution of sperm chemotaxis in the animal kingdom said that circumstantial to strong evidence exists in Hydrozoa (Colenterata), Ectoprocta, Cephalopoda Mollusca, Chaetognatha, Urochordata and Vertebrata. Pericentriolar processes in cnidarian (*Hydractinia*) spermatids described by M. G. Kleve and W. H. Clark, Jr. (USA) are apparently not involved in the direct spermatogenic events; possibly they have some role in the chemotactic behaviour of the cnidarian sperm. R. W. Atherton (USA) in his contribution submitted to ISRPI hypothesized that "neurochemical effects on invertebrate and vertebrate sperm motility may be active through a path linking acetylcholinesterase and adenylatecyclase activity".

Fertilizability can be induced in starfish oocytes by 1-methyladenine; mixing of germinal vesicle material with ooplasm is, however, necessary for genuine cleavage. Nuclei of sperm that enter an oocyte having intact germinal vesicle show no visible change until the germinal vesicle is made to break with 1-methyladenine. Sperm nuclei now swell indicating the involvement of the material of the germinal vesicle in the process. W. H. Clark, Jr. *et al.* (USA) presented evidence to show that the cortical rods in the cortex of mature oocytes in penaeids are composed of neutral mucopolysaccharides, and suggested that their discharge and dissipation may depend on Mg^{++} ions, a protease and possibly a phosphatase. D. F. Went (Switzerland) narrated his success with *in vitro* culture from oogonium to larva for several generations in the paedogenetic viviparous gall midge, *Heteropeza pygmaea*; determination of sex in the ovary depends on nutritive conditions for the feeding larvae.

The chemistry of sialoglycoproteins contained in the jelly coat substances of sea urchin eggs was presented by K. Hotta and M. Kurokawa (Japan); sialic acids in the sialoglycoprotein of *Pseudocentrotus depressus* consisted mainly of N-glycolyl- and N-aceto-glycolyl-4-methyl-4, 9-dideoxy neuraminic acids, whereas those of *Anthocidaris crassispina* and *Hemicentrotus pulcherrimus* were mostly N-glycolylneuraminic acid with some N-acetylyneuraminic acid. G. Anilkumar and K. G. Adiyodi (India) reported cyclicity in the secretory activity of the spermathecal glandular epithelium in the crab, *Paratelphusa hydrodromous* in relation to the vitellogenic cycle. This suggests that the spermathecal epithelium in crabs may be a target tissue of one or more of the crustacean reproductive hormones. T. S. Dhadialla (Kenya) presented the results of his investigations on the relative influences of mechanical and chemical factors of mating on engorgement in the female hard tick, *Rhipicephalus appendiculatus.* Results of Dhadialla seem to suggest that the stimuli for rapid engorgement in female ticks are mostly mechanical and partly chemical, the latter resulting from spermatophore and its contents.

Endocrine controls in ovarian maturation and the neural influences in oviposition received attention at ISRPI. T. S. Adams (USA) hypothesized that the oostatic hormone postulated by him in the housefly may regulate the release of neurosecretory material from the median neurosecretory cells. P. J. W. Olive (UK) presented evidence to show that prostomium is the source of a hormone that initiates and supports vitellogenesis in polychaetes. Spawning can be induced in gravid *Nephtys hombergi* if supra-oesophageal ganglion homogenate is administered; the homogenate of sub-oesophageal ganglion shows no such effect. K. Yamaoka (Japan) explained his studies on mechanisms releasing ovipositional behaviour in *Bombyx mori.* Copulation enhances egg-laying and also the spontaneous nervous activity of the isolated last abdominal ganglion (SMG IX) in female moths. Oviposition-stimulating substance contained in male sex secretions transferred into the female during coitus apparently affects Na^+ and K^+ permeability of the sheath of SMG IX; the change in ionic condition probably causes the activation of the ganglion.

The deliberations were interesting and useful and covered the structural, biochemical, histochemical, endocrinological, neurological, pharmacological, ecological, genetical and evolutionary aspects of reproductive function. An International Society for Invertebrate Reproduction (ISIR) was established at the Symposium. A 20-member executive council was elected with Wallis H. Clark, Jr. (USA) as the President, T. S. Adams (USA), B. R. Laurence (UK) and T. R. Odhiambo (Kenya) as the Vice-Presidents, K. G. Adiyodi (India) as the Secretary, P. J. W. Olive (UK) as the Joint Secretary and D. F. Went (Switzerland) as the Treasurer. Calicut symposium has been accepted as the First International Symposium on Invertebrate Reproduction of ISIR; the second is slated to be held in USA in 1978.

LETTERS TO THE EDITOR

ACCURATE DETERMINATION OF M 1–E 2 MIXING RATIO AND NUCLEAR STRUCTURE PARAMETER OF THE 39·58 keV TRANSITION IN 129 Xe

THE L– sub shell ratios of the 39·58 keV transition in 129 Xe were measured quite accurately by Geiger *et al.*[1], using an iron free double focussing beta ray spectrometer. The experimental values were used by Geiger *et al.*[1] to derive the values of the M 1–E 2 mixing ratio as M 1 + (0·075 ± 0·025) % E 2. Considering the low energy of the transition and the spins of the levels, namely, 3/2 and ½ which the transition connects the M 1 component should be l-forbidden. Consequently gamma transition matrix element should be smaller than in the normal case. As a result the anomaly parameter defined as the ratio of the penetration matrix element and the gamma ray matrix element should be sizeable. Such an anomaly factor was searched for, by Venkata Ramaniah *et al.*[2], by analysing the L subshell ratios of Geiger *et al.*[1], in conjunction with the theoretical tabulations of Hager and Seltzer[3,4] for conversion coefficients and penetration functions, in a bid to get an accurate value of the M 1–E 2 mixing ratio of this transition. A plot of λ *vs* δ^2 was made for experimental values of L_I/L_{II}, L_I/L_{III} and L_{II}/L_{III} in terms of the experimental conversion coefficient ratios and penetration functions. The centre of gravity of the area common to the three ratios would yield values of λ and δ^2. Since in Fig. 1, Venkata Ramaniah[2] derived the values of λ and δ^2 from a shaded area which is not common to the three subshell ratio lines, it was thought desirable to reanalyse the data from the point of view of mixing ratio and penetration parameter. In Fig. 1, we have given the diagram obtained from a careful analysis of experimental and theoretical data which is listed in Table I.

TABLE I

Experimental and theoretical subshell ratios used in the analysis

Sub shell Ratio	Theoretical		Experimental
	E 2	M 1	
L_I/L_{II}	0·0625	12·45	10· +0·625 − 0·577
L_{II}/L_{III}	0·7238	4·08	3·225 + 0·49 − 0·40
L_I/L_{III}	0·0452	50·8	32·258 + 4·2 −3·4

The values of δ^2 and λ are as under:

$$\delta^2 = (0\cdot04 \pm 0\cdot03)\ \%$$

$$\lambda = 12 \pm 6.$$

The sign of λ is compatible with the effective operator calculations of Subba Rao[5] which predict the sign of λ to be positive for all odd particle transitions.

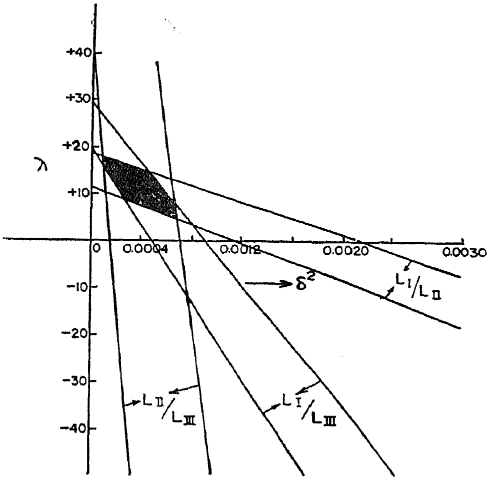

FIG. 1. Plot of λ *vs.* δ^2 for the 39·58 keV transition in 129 Xe.

The authors are thankful to Prof. B. S. Sood. for his constant encouragement and facilities. One of us (VSP) is grateful to the CSIR for financial help.

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C. S. KHURANA.

---

1. Geiger, J. S., Graham, R. L., Bergstrom, I., and Brown, F., *Nuc. Phy.*, 1965, **68**, 352.
2. Venkata Ramaniah, K., Seshi Reddy, T. and Vankata Reddy, K., *Curr. Sci.*, 1974, **43**, 406.
3. Hager, R. and Seltzer, E., *Nuc. Data*, 1968, **4 A**, 1.
4. — and —, *Ibid.*, 1969, **6 A**, 1.
5. Subba Rao, B. N., Private Communication.

X-RAY K-ABSORPTION EDGES OF Ge, GeO_2 AND Se, SeO_2

It is known that in a solid, the valence state electrons participate in the chemical bonds. Several things happen[1] when the chemical composition of the solid is varied. The combined effect will be to shift the energy of inner state X-ray absorption edge as the chemical bonds around an atom in a solid are changed. The purpose of this note is to report such shifts when germanium and selenium form oxides.

A Machlett sealed X-ray tube with a tungsten target at 12 mA and 15 kV has been used to record the photographs. A 40 cm Cauchois type bent crystal spectrograph was used as analyser with $(\bar{2}01)$ mica reflection planes. The resolution of the spectrograph was such, as to record a resolved spectrum of Mo $\beta_{1,3}$ doublet in first and second order. Absorption edges were recorded on kodak X-ray films in exposures lasting from 6 to 8 hours. Microphotometer records were obtained with a magnification of × 4. The dispersion was ~ 12 x.u./mm in the region of discontinuity. The absorption screens were prepared by uniformly spreading the substances, in the powder form on a scotch tape and covering it with another piece of the tape. The following tungsten reference lines were used for measurements : $L\gamma_1$, $L\gamma_2$, $L\gamma_3$, $L\gamma_4$ and $L\gamma_5$.

Germanium metal powder supplied by Messrs. Johnson Mathey, Co. Ltd., London and selenium powder supplied by Messrs. Shinko Chemical Co. Ltd., Japan, were used. Germanium dioxide and selenium dioxide were obtained from National Chemical Laboratory, Poona, India.

The results of our measurements on the microphotometer records with magnification, × 4 are given in the Table I. The electronic configuration of germanium is $3d^{10}\ 4s^2\ 4p^2$. In germanium the covalent bonding is due to sp^3 hybrid formation.

Table I

K-absorption edge of Ge, GeO_2, Se and SeO_2

Substances	λ in x.u. ± 0·05	ν/R	$\Delta\nu/R$	Δ eV ± 0·3	Remark
Ge	1114·34	817·76	0·00	0·00	..
GeO_2	1114·26	817·86	+ 0·1	+ 1·35	..
Se	977·60	932·15	0·00	0·00	..
SeO_2	976·71	933·00	+ 0·85	+ 11·53	W.L.

It leads to an energy band model having a small energy gap, 0·72 eV. In germanium intrinsic conductivity is, therefore, observed at room temperature. The large carrier drift mobility in germanium suggests that the valence electrons have a tendency to resemble free electrons. Hence, as expected, a white line has not been observed. The shift in GeO_2 is 1·35 eV. A larger shift was expected on the basis of covalent character. The percentage of covalency as calculated by Suchet's formula[2] is only 31. However, a large shift has not been observed in GeO_2 as reported by Sapre and Mande[3]. In crystalline GeO_2 the contribution to the covalency is expected to be large due to the bonds in the crystal[4]. This would result in smaller shift in GeO_2. The situation may be compared with that of Ga_2O_3 where it was found that the covalent nature of the bond results in small shift[5,6].

It is seen that selenium metal edge, shifts towards the short wavelength side, in the selenium dioxide (SeO_2) by 11·53 eV with respect to the pure selenium. A marked white line (WL) in the case of SeO_2 has been observed. Although selenium is a semiconductor, no predominant white line has been observed in this case. White line has been reported by DesLattes and deBen[7] in selenium spectrum using very thin absorbing screens obtained by vacuum evaporation technique. Other studies[8] have established that white line near the absorption edge disappears gradually by the use of increasingly thicker screens. Difficulty of obtaining white line in selenium using powder absorbing screens has also been pointed out by Mande *et al.*[9]. The valency of selenium in SeO_2 is 4 and therefore large shift as expected has been observed[6].

Author is grateful to Dr. B. K. Agarwal of Physics Department, Allahabad University, Allahabad, for interest in this work.

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---

1. Verma, L. P. and Agarwal, B. K., *J. Phys. C.*, 1968, 1 (2), 1658.
2. Suchet, J. P. and Bailly, F., *Ann. Chim.*, 1965, 10, 517.
3. Sapre, V. B. and Mande, C., *Indian J. pure and appl. Phys.*, 1974, 12, 74.
4. Syrkin, Y. K. and Dyatikina, M. E., *Structure of Molecules and the Chemical Bonds*, Dover Pub., N.Y., 1964, 330, 331.
5. Verma, L. P. and Agarwal, B. K., *Indian J. pure and appl. Phys.*, 1967, 5, 616.
6. Agarwal, B. K. and Verma, L. P., *J. Phys. C.*, 1970, 3, 535.
7. Deslattes, R. D. and DeBen, H. S., *Phys. Rev.*, 1959, 115, 71.
8. Parrat, L. G., Hampstead, C. E. and Jossem, E. L., *Ibid.*, 1957, 105, 1228.
9. Mande, C., Patil, R. N. and Nigaveker, A. S., *Indian J. pure and appl. Phys.*, 1965, 3, 483.

THE PETROGRAPHIC STUDIES AND THE AGE DETERMINATION OF KOIDAL GNEISS, KUMAON HIMALAYA

THE age and origin of granitic and migmatitic rocks, which cover a large tract in the Kumaon Himalaya, have been a subject of study by many workers and widely different views have been expressed on this subject. For instance, the gneissic rocks of Ramgarh area of Kumaon Himalaya were termed as quartz porphyries by Heim and Gansser[1] and were assigned a Pre-Cambrian age. On the other hand, Pande[2] designated these rocks as migmatites on the basis of field and petrographic studies. He assigned a metamorphic origin and Tertiary age to these rocks. Isotopic age data can provide useful information for resolving the question of age and even of origin of these rocks. The present communication reports the first Rb–Sr whole rock study for the gneissic rocks of Koidal area, District Nainital, Kumaon Himalaya. A brief account of the field-observations and petrographic studies is also given.

The rock varies in appearance from streaky gneiss at one end to subfoliated granite at the other. In general, it has a conformable contact with the surrounding chlorite-sericite schist. Under thin section, the felspar phenocrysts are usually microcline. Fine grains of quartz, chlorite and sericite occur in the ground mass. Parallel to sub-parallel arrangement of the flaky minerals gives it a foliated appearance. Fracturing of the phenocrysts of microcline and some straining-effect in the quartz-grains provide evidence of the rock having been involved in a tectonic event. On the whole, the gneissic rocks under study, on the basis of lithological similarities and tectonic setup, can be correlated to the foliated migmatites of Ramgarh area in the West (Pande[2]) and Debguru gneiss of Gorakhnath formation of Lohaghat group in the East (Mishra and Sharma[3]). However, the outcrop width of the gneissic rocks increases from about ½ to 1 km in the Ramgarh to about 3 km in the Koidal area and to more than 5 km in the Debguru area.

Rb–Sr studies for the Koidal area have been made by use of conventional mass-spectrometric and isotopic-dilution techniques. In conformity with our earlier work[4,5], the decay constant for rubidium has been taken as $1{\cdot}47 \times 10^{-11}$/year and the initial $Sr^{87}/Sr^{86} = 0{\cdot}709$. The results of analyses are given below: Sample no. and location = PHS/108 (29° 22′ 15″; 79° 42′ 15″); (Sr^{87}/Sr^{86}) total $= 0{\cdot}854 \pm 0{\cdot}015$; $Rb^{87} = 100{\cdot}4$ ppm; Common Sr = 121 ppm; Apparent age = 1170 ± 120 m.y.

The above results point out that the Koidal gneiss is of Pre-Cambrian age. The isochron age may, however, turn out to be younger than the above-mentioned apparent age.

Shri D. K. Chadha, Shri J. K. Bhalla and Shri J. S. Gill gave assistance in the early phases of this work. The authors' thanks are due to the Bhabha Atomic Research Centre, Bombay, for loaning of the mass-spectrometer. The financial assistance of the C.S.I.R. and INSA are gratefully acknowledged.

Physics Department, V. B. BHANOT.
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Chandigarh, V. P. SINGH.
July 18, 1975. A. K. GOEL.

---

1. Heim, and Gansser, *Central Himalaya, Geological Observations of Swiss Expedition*, 1939.
2. Pande, I. C., *Jour. Sci. and Res. B.H.U.*, 1956, 7, 88.
3. Misra, R. C. and Sharma, R. P., *Jour. Geol. Soc. Ind.*, 1967, 8, 110.
4. Bhanot, V. B., Gill, J. S., Arora, R. P. and Bhalla, J. K., *Curr. Sci.*, 1974, 43, 208.
5. —, Goel, A. K., Singh, V. P. and Kwatra, S. K., *Ibid.*, 1975, 44, 219.

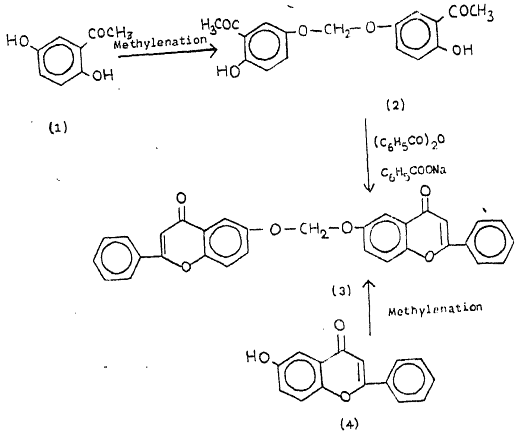
STUDIES IN METHYLENATION OF SOME POLYPHENOLS; ISOLATION OF DIMERS LINKED THROUGH METHYLENEDIOXY GROUP

METHYLENATION of quinacetophenone and 6-hydroxy flavone yielded the bimolecular compounds linked through a methylenedioxy group in 5 and 6 position respectively. The dimer obtained from quinacetophenone could be converted to that obtained from 6-hydroxy flavone through cyclisation using benzoic anhydride and sodium benzoate.

Methylenation, regarded as a biogenetic process, has been commonly reported in plant polyphenolics[1-5]. In the past several years methylenations have been carried out in polyphenols in the laboratory, *e.g.*, in 5, 6, 7, 4′-tetrahydroxyisoflavone[6] and Vogoletin[7] methylenation of a catechol unit in 6, 7- positions was successful. Methylenation of 7-hydroxyflavonoids and carbonyl derivatives of resorcinol has been reported to yield bimolecular compounds linked through a methylenedioxy group in the reactive 7 and 4 positions respectively[8].

During the present work quinacetophenone (1) has been methylenated with methylene iodide in presence of potassium carbonate and the isolated product was found to be bi-(2-hydroxy-1-acetophenonyl-2-oxy) methane (2).

Condensation of compound (2) with benzoic anhydride and sodium benzoate yielded bi-(6-flavonyloxy) methane (3). This was found to be identical with the product obtained by the methylenation of 6-hydroxyflavone (4), by mixed melting point, co-chromatography and comparison of their infrared spectra.



EXPERIMENTAL

Bi-(2-hydroxy-1-acetophenoyl-5-oxy) methane.—Quinacetophenone (2 g) was refluxed in acetone (40 ml) with methylene iodide (1 ml) and anhydrous potassium carbonate (10 g) for 4 hrs. Acetone was distilled off and water (100 ml) added to the residue. The solid, after filtration and drying, crystallized from ethanol as colourless needles, m.p. 192–193° ν_{max} 1630 cm^{-1} (H–bonded C=O). It gave positive methylenedioxy test (found C, 64·45; H, 5·0, $C_{17}H_{16}O_6$ required C, 64·6; H, 5·1%).

Synthesis of bi-(6-flavonyloxy) methane :

(1) *from bi-(2-hydroxy-1-acetophenonyl-5-oxy) methane.*—A mixture of bi-(2-hydroxy-1-acetophenonyl-5-oxy) methane (2·6 g) benzoic anhydride (35 g) and sodium benzoate (7 g) was heated at 180–185° for 4 hrs. It was then cooled and the pale brown mass was broken up and added to alcohol (170 ml). The mixture was boiled during the gradual addition of potassium hydroxide (18 g) dissolved in 40 ml of water and then heated for 30 minutes under reflux. The alcohol was removed by distillation. The residue was dissolved in water. The filtered solution was saturated with carbon dioxide. The precipitate was filtered, dried and then crystallised from ethyl acetate as colourless microprisms, m.p. 236–237°. R_f = 0·65 chloroform-ethyl acetate (3 : 20) ν_{max} 1640 cm^{-1} (flavone C=O (found C, 75·95; H, 4·0; mol. wt. (Rast method), 469; $C_{31}H_{20}O_6$ requires C, 76·21; H, 4·1%; mol. wt., 488).

(2) *Dimer from 6-hydroxyflavone.*—6-hydroxyflavone (0·2 g), acetone (125 ml), methylene iodide (0·1 ml) and anhydrous potassium carbonate (4 g) were refluxed together for 40 hrs. Acetone was distilled off and water (100 ml) was added to the residue. The solid (0·1 g), after filtration and drying, crystallized from ethyl acetate as colourless microprisms, m.p. 236–237°. Mixed m.p. with the compound synthesised by the method (1) was not depressed. It gave positive methylenedioxy test. R_f = 0·65; chloroform-ethyl acetate (3 : 20) [found C, 76·1; H, 4·1; mol. wt. (Rast method), 471; $C_{31}H_{20}O_6$ requires C, 76·21; H, 4·1%; mol. wt., 488].

Methylenedioxy test.—The methylenedioxy test was performed by heating the compound with gallic acid and conc. H_2SO_4 on a boiling water bath. Emerald green colour was taken as a positive reaction.

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India, *June* 10, 1975.

---

1. Gopinath, K. W., Kidwai, A. R. and Prakash, L., *Tetrahedron,* 1961, 16, 201.
2. Cooker, W., Dahl, T., Dampsey, C. and McMurry, T. B. H., *Chem. and Ind.,* 1962, p. 216.
3. Van Duuran, B. L., *J. Org. Chem.,* 1961, 26, 5013.
4. Birch, A. J., Moore, B. and Mukerjee, S. K., *Tetrahedron Letters,* 1962, 15, 673.
5. Khanna, R. N. and Seshadri, T. R., *Tetrahedron,* 1963, 19, 219.
6. Arora, S. K., Jain, A. C. and Seshadri, T. R., *Ibid.,* 1962, 18, 559.
7. Rangaswami, S. and Rao, H., *Proc. Indian Acad. Sci.,* 1959, 49 A, 241.
8. Grover, S. K., Jain, A. C. and Seshadri, T. R., *Tetrahedron,* 1964, 20, 555.

EFFECT OF ELECTROLYTES ON THE RATE OF HYDROLYSIS OF 1-PHENYLNEOPENTYL CHLORIDE IN WATER

THE diagnostic introduced by Ingold[1] of adding a lyate ion or another nucleophile for distinguishing between S_N1 and S_N2 mechanisms has been criticised by Hine[2]. Following this, this technique has either been ignored[3] or misinterpreted[4]. However, Schleyer has recently proved the correctness of Ingold's view, so far as azide ions are concerned[5]. We have now studied a system, which is not readily susceptible to S_N^2 attack, using the less nucleophilic bromide as the reagent.

1-phenylneopentyl chloride has been shown to solvolyse by the S_N^1 mechanism, even in relatively non-ionizing solvents[6,7]. It has also been shown that its structure disfavours S_N^2 attack[6]. We have studied the kinetics of hydrolysis in solvent water in which, because of the high ionizing capacity of the medium, an S_N2 pathway is even less likely Our results (Table I) show that non-common ion electrolytes show negligible ionic strength effect, which is consistent with the Debye–Hückel theory. Addition of bromide ions on the other hand shows marked increase in rate; the non-linearity between the increase in rate and the bromide ion concentration clearly indicates that its effect is not due to the normal type of salt effect described by Winstein[8]. Further support for this conclusion comes from the observation that the rate constants in the presence of bromide ions increase with the progress of reaction. This effect is not seen either in the absence of bromide ions or in the presence of other non-common ion electrolytes.

TABLE I

Effect of added ions on the rate of hydrolysis of 1-phenylneopentyl chloride in water

Temp. 45·70° C (RC1) = 0·0006 M

Added salt	Concentration (M)	$10^4 k_1$ (s^{-1})	% increase in rate due to added salt
Nil	..	1·47	..
$LiClO_4$	0·05	1·47	0·0
$NaNO_3$	0·05	1·52	2·7
NaBr	0·01	1·63	10·9
,,	0·05	2·04	38·8
,,	0·10	2·15	46·3

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1. Bateman, L. C., Hughes, E. D. and Ingold, C. K., *J. Chem. Soc.*, 1940, p. 960.
2. Hine, J., *Physical Organic Chemistry*, Second Edition, McGraw-Hill, Book, Co., Inc., Tokyo, 1962, p. 131.
3. Sneen, R. A. and Larsen, J. W., *J. Amer. Chem. Soc.*, 1969, **91**, 362.
4. Senatore, L., Sagramora, L. and Ciuffaril, E., *J. Chem. Soc.*, Perkin II, 1974, p. 722.
5. Rafer, D. J., Harris, J. M., Hall, R. E. and Schleyer, P. V. R., *J. Amer. Chem. Soc.*, 1971, **93**, 4821.
6. Winstein, S. and Morse, B. K., *Ibid.*, 1952, **74**, 1133.
7. Anantaraman, R. and Saramma, K., *Tetrahedron*, 1965, **21**, 535; *Canad. J. Chem.*, 1965, **43**, 1770.
8. Fainberg, A. H. and Winstein, S., *J. Amer. Chem. Soc.*, 1956, **78**, 2780.

THE OBSERVED ACTIVATION OF ENZYMES BY MN++ IS SOMETIMES AN ARTIFACT

DURING a study of the effect of metal ions on the activity of a bacterial malic dehydrogenase (EC 1.1.1.37), it was found that Mn^{++} at concentrations as low as 10^{-5} M stimulated the rate at which absorption at 340 nm increased when malate and NAD were in excess. When oxaloacetate (OAA) and NADH were in excess, addition of Mn^{++} reduced the rate at which absorption at 340 nm decreased (Fig. 1). The kinetics of these reactions suggested the presence of a contaminating activity such as malate-NAD-oxidoreductase (EC 1.1.1.38). This enzyme, which is activated by Mn^{++}, catalyzes the conversion of malate to pyruvate and carbon dioxide[1].

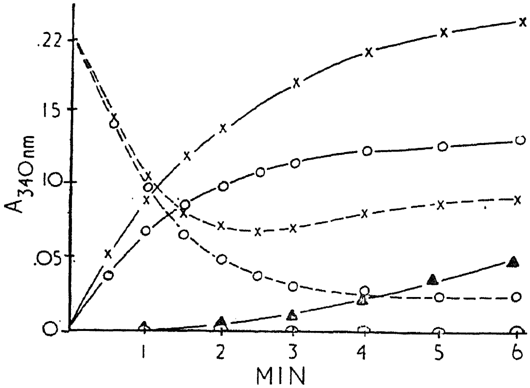

FIG. 1. Effect of Mn^{++} on a malic dehydrogenase (EC 1. 1. 1. 37). Assays were done spectrophotometrically at 340 nm. Dotted lines: enzyme with oxaloacetate and NADH in tris buffer at pH 8·5. Solid line with circular symbols: enzyme with malate and NAD at pH 8·5. Curves with X symbols contained in addition 1 mM Mn^{++}. Curve with triangles: buffer only *plus* 1 mM Mn^{++} at pH 8·5 Base line (half circles): buffer at pH 7·0 *plus* 1 mM Mn^{++}, or buffer at pH 8·5 with 1 mM EDTA *plus* 1 mM Mn^{++}, or buffer alone.

We sought direct evidence for the presence of this enzyme by testing for the formation of both pyruvate and CO_2, the latter was determined manometrically after acidification. The CO_2 resulted from the spontaneous decomposition of OAA and its amount was independent of the Mn^{++} concentration. What pyruvate was detected by gas-liquid chromatography (GLC) was also unrelated to metal ions and was traced, in part, to the breakdown of OAA during methylation for GLC. Pyruvate was also estimated with lactic dehydrogenase; no significant increase could be attributed to the presence of Mn^{++} in the malic dehydrogenase assay. Further protein purification steps did not increase the specific activity of the Mn-dependent activity, nor did it alter the fact that Mn^{++} stimulated the rate of reaction in one direction but not in the other. This suggested a non-enzymatic reaction. Careful examination of the system showed that the absorbancy of solutions of Mn^{++} in buffer at pH 8·5 increased steadily with time at a rate of the order of that expected for enzymic preparations. This effect was obviated by pH values below 7·0 or in the presence of ethylenediaminetetracetic acid (EDTA) as shown in Fig. 1. The rate of change in absorption was dependent on the pH and also the presence of dissolved oxygen. The reaction is, in fact, the oxidation of Mn^{++} to Mn^{+4}. The product, which is probably largely $Mn(OH)_4$ or a mixture of basic salts, is soluble at low concentrations. The solutes become colloidal and finally precipitate particularly at higher initial Mn^{++} concentrations and at higher pH values. A fairly large number of enzymes are activated by Mn^{++}. We believe, however, that some reports of stimulation by Mn^{++} (but not by other metal ions) should be re-examined. For example the NAD-linked glucose dehydrogenase of *Bacillus cereus* is reported activated at pH 8 by 10^{-4} M Mn^{++}. Other metals had no effect. Stimulation by Mn^{++} is demonstrated, but the term—activation—which is used *may* be misleading[2]. Was the appropriate control (Mn^{++} in buffer alone) run ? It is extremely important that it should be particularly with spectrophotometric assays at wavelengths below about 450 nm at pH 7 or above, in the presence of air.

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1. Kaufman, S., Korkes, S. and delCampillo, A., *J. Biol. Chem.*, 1951, 192, 301.
2. Sadoff, H. L., In : *Methods in Enzymology* (W. A. Wood, ed.), Academic Press, New York, 1966, 9, 103.

CYCLIC NUCLEOTIDE PHOSPHODIESTERASE ACTIVITY IN *CICER ARIETINUM*

CYCLIC 3 : 5 adenosine monophosphate (cAMP) mimics the action of indol-yl-3-acetic acid (IAA) in stimulating the activity of tryptophan oxygenase in *Cicer arietinum*[1]. Furthermore, exposure of seedlings to IAA leads to an increase in the adenyl cyclase activity[2]. cAMP also stimulates RNA and protein synthesis[3] and protein phosphorylation[4] in the seedlings. These findings suggest a regulatory role for cAMP in germination[5] and it was of interest, therefore, to examine the activity of cAMP phosphodiesterase which mediates the hydrolysis and thereby controls the intracellular concentration of cAMP in living cells. cAMP phosphodiesterase activity[6] was present in dormant seeds and registered a significant rise after 72 hr germination. The enzyme prepared from 72 hr seedlings (whole seeds *miuns* testae and cotyledon) was localized in the supernatant fraction recovered after sedimenting organelles at 100,000 g. The enzyme had an optimum pH around 5 and hydrolysed besides cAMP, cyclic uridine monophosphate, cyclic guanosine monophosphate and dibutyryl cAMP. Theophylline or theobromine in a concentration range of 100 μM–1 mM had no inhibitory effect and imidazole (100 μM) or IAA up to 1 mM had no activating effect on the enzyme. The plant enzyme thus seems to be different from mammalian cyclic AMP phosphodiesterases.

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1. Azhar, S. and Krishna Murti, C. R., *Ind. J. Biochem. and Biophys.*, 1971, 8, 210.
2. — and —, *Biochem. Biophys. Res. Commns.*, 1971, 43, 58.
3. Srivastava, A. K., Azhar, S. and Krishna Murti, C. R., *FEBS Letters*, 1973, 33, 239.
4. —, — and —, *Int. Conference on Cyclic AMP*, Vancover, July 1974.
5. —, — and —, *FEBS Letters*, 1974, 47, 330.
6. Butcher, R. W. and Sutherland, E. W., *J. Biol. Chem.*, 1962, 237, 1244.

OCCURRENCE OF AVICULARIN IN THE LEAVES OF *CINCHONA OFFICINALIS* AND *C. ROBUSTA*

THE genus *Cinchona* belonging to the family Rubiaceae and well-known for its alkaloids, has not been studied in any detail for the presence of polyphenols. We have earlier reported[1] the occurrence of mannitol in twenty plants and flavonoids in seven members of this family. The isolation of reynoutrin (a compound misidentified earlier as a xyloside[2] and later shown to be an arabinoside[3]) from *Cinchona ledgeriana*[4] has been reported. In view of these it was considered desirable to examine the leaves of

C. officinalis and *C. robusta* grown in South India and our findings are recorded below.

Adopting the standard procedure for the isolation of flavonoids, the aglycones present in the leaves were found to be mainly quercetin with small quantities of kaempferol. Of the three glycosides present, the one in appreciable quantity was isolated in pure form as light yellow needles, m.p. 210–12° (decomp). It was purple under U.V. changing to yellow with NH_3 and had λ_{max}. (nm) 259, 359 and λ_{min} 238, 286 (EtOH) and showed shifts with diagnostic reagents[5] characteristic for a quercetin-3-0-glycoside. Its IR spectrum (KBr) exhibited bands at 1650, 1600, 1500, 1360, 1195, 1110, 1040, 1000, 945, 920; 825 and 800 cm^{-1}. On acid hydrolysis it yielded quercetin and L–arabinose in equal proportion. The identity of the sugar as L–arabinose was established by co-chromatography of the sample with authentic arabinose and xylose. They had R_f (arabinose, xylose) : 0·27, 0·27 (BAW) ; 0·55, 0·50 (phenol); 0·45, 0·48 (*t*-BAW) ; 0·30, 0·35 (ethyl acetate : pyridine : water = 10 : 4 : 3) ; 0·25, 0·27 (butanol : ethanol : water = 4 : 1 : 1) and 0·36, 0·44 (benzene : butanol : pyridine : water = 1 : 5 : 3 : 3). Thus, the glycoside was identified as quercetin–3–0–L–arabinoside and a direct comparison, co-chromatography and superimposable I.R. spectra with an authentic compound showed it to be avicularin (quercetin–3–0–α–L–arabinoside).

In order to ascertain whether the quercetin glycoside isolated from *C. ledgeriana* was really quercetin–3–xyloside or the so far misidentified reynoutrin, an attempt made to procure a small quantity of the sample isolated from *C. ledgeriana*[4] for comparison was unsuccessful. The absence of record of the occurrence of quercetin–3–0–xyloside so far in nature, the earlier misidentification of reynoutrin as a xyloside and the present isolation of avicularin from *C. officinalis* and *C. robusta* warrant a re-examination of the true nature of the quercetin pentoside[4] isolated from *C. ledgeriana*[4].

Our grateful thanks are due to Shri S. Kalyanasundaram, Botanist, Government Cinchona Plantations, Dodabetta, Nilgiris, for authentic leaves of *C. officinalis* and *C. robusta,* Drs. T. R. Govindachari and B. S. Joshi, Ciba-Geigy Research Centre, Bombay–63, and Dr. L. Horhammer, University of Munich, Germany, for the spectral data and the Principal, JIPMER, for encouragement.

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---

1. Subramanian, S. S. and Nair, A. G. R., *Phytochem.*, 1971, 10, 2125.
2. Nakaoki, T. and Morita, N., *J. Pharm. Soc.*, Japan, 1956, 76, 323.
3. Horhammer, L. *et al.*, *Tetrahedron Letters*, 1966, p. 567.
4. Dhar, D. N. and Munjal, R. C., *Curr. Sci.*, 1974, 43, 479.
5. Mabry, T. J., Markham, K. R. and Thomas, M. B., *The Systematic Identification of Flavonoids,* Springer Verlag, Berlin, 1970.

THE EFFECT OF TOLUENE, PHENOBARBITAL AND 3-METHYLCHOLANTHRENE ON HEPATIC MICROSOMAL LIPID PEROXIDATION

Phenobarbital and 3-methylcholanthrene are known to be potent inducers of a range of hepatic microsomal xenobiotic metabolizing enzymes. It has been reported that the pretreatment of rats with either of these compounds produces inhibition of hepatic microsomal lipid peroxidation[1]. Numerous attempts have been made to study the levels of liver lipid peroxides in carbon tetrachloride treated rats[2-4] and it has been reported that the butylated hydroxytoluene[5,6] and benzene[7] significantly reduce the lipid peroxide formation.

Except for a preliminary study[8], involving some aromatic hydrocarbons, there is no recorded information about the effect of organic solvents on hepatic microsomal lipid peroxidation in rat. Accordingly, the present paper reports the effect of toluene on liver microsomal lipid peroxidation. Furthermore, the effect of pretreatment of phenobarbital and 3-methylcholanthrene on the toxicity of toluene was also studied.

Materials and Methods

Hindustan antibiotic strain adult male (200–250 gm) and female (135–160 gm) albino rats were obtained from Hindustan-Antibiotics, Poona. The animals were kept in an air-conditioned room and supplied with rat pellets (obtained from Hindustan-Lever Ltd., Bombay) *ad libitum* prior to the initiation of the experiments. All animals were fed on a synthetic diet for 1 week before initiation of the experiments. The composition of the diet was as reported earlier[9] except that the casein content was increased to 18%. The rats were then allotted to the following groups and pair fed during the experimental period. (1) Control, (2) Toluene treated, (3) Phenobarbital treated, (4) Phenobarbital and toluene treated, (5) 3-Methylcholanthrene treated and (6) 3-Methylcholanthrene and toluene treated. Toluene (0·72 ml/kg body wt.) was administered orally to the rats using corn oil as a carrier in the morning (before feeding) for 2 successive days. Sodium phenobarbital (80 mg/kg body wt.) was injected intraperitoneally, daily, in the morning between 8·0–9·0 a.m. for 3 days, Rats in group 4 which were treated with phenobarbital were further treated with toluene for 2 successive days. 3-Methylcholanthrene (obtained from Calbiochem, Switzerland) was injected intraperitoneally at a dose of 20 mg/kg body wt. in corn oil in the morning for 2 days. The rats in group 6 which were treated with 3-methylcholanthrene were further treated with toluene for 2 successive days.

Tissue Preparation.—The animals were killed by decapitation, 24 hours after the last injection. The whole livers were carefully perfused with 0·9% ice-cold saline, excised, weighed, minced and homogenized (1 : 4 w/v) in ice-cold 50 mM Tris-HCl buffer, pH 7·4 containing 1·15% KCl. All tissue preparations were made at – 2° C. The microsomes were isolated by the procedure of Baker *et al.*[10]. The microsomal protein was determined by the biuret method[11] using crystalline bovine serum albumin as the standard.

Lipid Peroxidation.—NADPH linked lipid peroxidation was assayed according to the method of Ernster and Nordenbrand[12]. Ascorbate-induced lipid peroxidation was carried out in the same medium but nicotinamide was omitted and NADPH was replaced by 1 mM ascorbate. The malonaldehyde formation was measured by the thiobarbituric acid reaction[13].

Results and Discussion

Table I shows the effect of various treatments on lipid peroxidation. Phenobarbital and 3-methylcholanthrene treatment resulted in lowering the NADPH linked and ascorbate induced lipid peroxidation irrespective of the sex. The magnitude of decrease in 3-methylcholanthrene treated male rats was more than in phenobarbital treated ones. Phenobarbital and 3-methylcholanthrene treatment resulted in a similar reduction in lipid peroxidation in female rats.

Toluene treatment lowered both the NADPH linked and ascorbate-induced lipid peroxidation irrespective of sex ; however, the decrease was more in male as compared to female rats. Lipid peroxide formation was further decreased in rats pretreated with phenobarbital prior to the administration of toluene in both sexes. However, pretreatment of 3-methylcholanthrene prior to the administration of toluene reduced lipid peroxidation significantly only in the male rats.

The inhibitory effect of toluene on lipid peroxidation may be due to the antioxidant property of toluene metabolites. Further decrease in lipid peroxidation in rats pretreated with phenobarbital or 3-methylcholanthrene may be due to competition for a common flavoprotein cytochrome *c* reductase[1] for the metabolism of these drugs. These results agree with the report of Wills[7].

TABLE I

*Effect of treatment of toluene on NADPH linked and ascorbate induced microsomal lipid peroxidation with and without pretreatment of phenobarbital and 3-methylcholanthrene in adult male and female rats**

Group	NADPH-linked lipid peroxidation†		Ascorbate-induced lipid peroxidation†	
	Male	Female	Male	Female
Control	16·20 ±0·20	27·40 ±0·20	17·80 ±0·20	19·40 ±0·20
Toluene treated	13·80 ±1·40^a	25·80 ±0·20^a	13·95 ±0·85^b	17·40 ±0·20^a
Phenobarbital treated	14·00 ±0·40^a	17·20 ±0·40^c	14·80 ±0·40^b	16·20 ±0·20^b
Phenobarbital + toluene treated	13·00 ±0·20^a	19·00 ±0·20^b	12·68 ±0·20^a	17·80 ±0·20^a
3-Methylcholanthrene treated	11·20 ±1·60^c	21·20 ±1·20^b	12·20 ±0·20^c	17·00 ±1·00^a
3-Methylcholanthrene + toluene terated	8·40 ±0·40^c	21·20 ±0·40^b	9·20 ±0·40^c	16·80 ±0·80^a

* The results are mean values (±SEM ; 10 rats in each group).

† Activity is expressed as *n*-moles malonaldehyde formed/min/mg protein.

^{a} = $P < 0{\cdot}05$; ^{b} = $P < 0{\cdot}01$; ^{c} = $P < 0{\cdot}001$.

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1. Orrenius, S., Dallner, G. and Ernster, L., *Biochem. Biophys. Res. Comm.*, 1964, 14, 329.
2. Comporti, M., *Boll. Soc. Ital. Biol. Sper.*, 1965, 41, 222.
3. Priest, R. E., Smuckler, E. A., Iseri, O. A. and Beneditti, E. P., *Proc. Soc. Exptl. Biol. Med.*, 1962, 111, 50.
4. Recknagel, R. O. and Ghoshal, A. K., *Exp. Mol. Pathol.*, 1966, 5, 108.

5. Sgaragli, G. and Conti, M. R., *Pharmacol. Res. Comm.*, 1971, 3, 315.
6. Vainio, H., *Res. Comm. Chem. Pathol. Pharmacol.*, 1974, 2, 289.
7. Wills, E. D., *Biochem. J.*, 1969, 113, 333.
8. Pawar, S. S. and Mungikar, A. M., *Ind. J. Biochem. Biophys.*, 1975, 12, 133.
9. Patel, J. M. and Pawar, S. S., *Biochem. Phtrmacol.*, 1974, 23, 1467.
10. Baker, R. C., Coons, L. B. and Hodgson, E., *Chem. Biol. Interactions*, 1973, 6, 307.
11. Gornall, A. G., Bardawill, C. J. and David, M. M., *J. Biol. Chem.*, 1949, 177, 751.
12. Ernster, L. and Nordenbrand, K., *Methods Enzymol.*, 1967, 10, 574.
13. Bernheim, F., Bernheim, M. L. and Wilbur, K. M., *J. Biol. Chem.*, 1948, 174, 257.

ON THE OCCURRENCE OF PRIAPULIDS IN THE LITTORAL REGION OF BHIMILIPATNAM (VISAKHAPATNAM, BAY OF BENGAL)

VERY little is known about the eucoelomate phylum Priapulida, which are marine vermiform cylindrical organisms attaining a length of over 12 cm with a bulbous anterior end, destitute of appendages and with an eversible proboscis. Surface of the body is superficially segmented and covered with spines.

Priapulids are hitherto known to occur only in the high latitudes and it is agreed that they have bipolar distribution and are absent in the tropics and subtropics[1-3]. Of late they have been reported from the Red Sea, Bermuda and Curacuo as meiofaunal components[4-6]. It is interesting to note that these organisms wherever reported were based on a small number of specimens only, whilst Coul[5] observed them as an important faunule constantly occurring in the meiobenthos forming 4·7% of the total proportion and ranked fourth in abundance accounting for 2% of all the meiobenthic animals collected. However, these animals were not known from Indian Ocean excepting for the Red Sea record. In the present communication the authors report on the occurrence of priapulids for the first time from the peninsular India and for the second time from Indian Ocean.

While engaged in the studies of littoral benthos of a backwater region at Bhimilipatnam about 35 km north of Visakhapatnam (70° 44′ N and 83° 23′ E) on the east coast of India, two priapulids (Fig. 1) were collected from a sediment comprising mud with sand, rock and shell pieces and rich in organic debris. The variation of ambient salinity ranged from 10‰ (during monsoon) to 37‰ (during summer) and temperature from 20° to 35° C.

Absence of tentacles, eversible proboscis with longitudinal rows of papillae, superficially segmented body covered with spines, mouth associated with recurved spines are the salient features of the specimens collected.

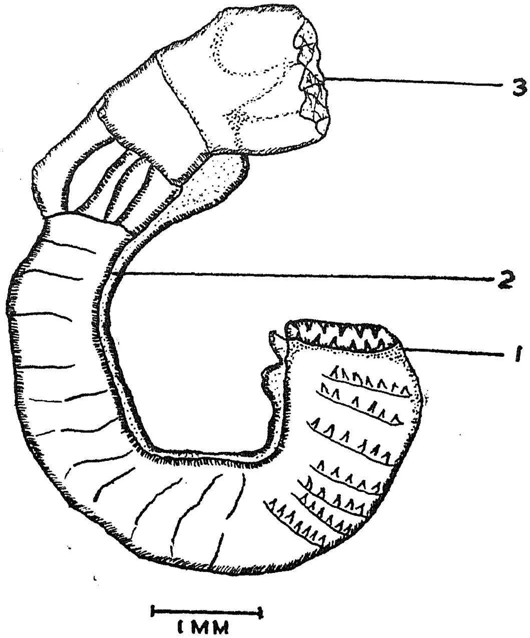

FIG. 1. 1, Introvert partially extruded showing the hooks; 2, Ventral fold; 3, Caudal region.

The present record of the occurrence of priapulids from a tropical location, further extends their traditional habit and distribution into the tropical Indian Ocean region. Thus the hitherto accepted bipolar distribution of these organisms appears to be no more valid.

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1. Baltzer, V. F., *Handbuch Der Zoologie II* (9) (Walter De Gruyter and Co.), 1931.
2. Shapeero, W. L., *Science*, 1961, 133 (2345), 879.
3. —, *Amer. Midland. Nat.*, 1962, 68, 237.
4. Remane, A., "The systematic position and phylogeny of pseudocoelomates," in E. C. Dougherty *et al.*, ed., *The Lower Metazoa*, Comp. Biol. Phylo., Univ. Calif. Br. Berkeley, 1963, p. 247.
5. Coul, B. C., *Thesis*, Lehigh. Univ., 1968.
6. Van der Land, J., *Zoologishe. Medeoilimgen.*, 1968, 42 (22), 237.

A STUDY ON THE PATTERN OF EXCRETION IN THE FRESH WATER BIVALVE *LAMELLIDENS CORRIANUS*

COMPARATIVELY very little attention has been focussed on nitrogen metabolism in lamellibranch molluscs[1,2]. The available data indicate that the marine bivalves are ammonotelic with lesser amounts of urea and free amino acids also contributing to the total nitrogen of the urine. Since only scant data are available on this aspect in fresh water forms[3], it was thought that an investigation on the pattern of excretion in the common fresh water mussel, *Lamellidens corrianus,* would yield useful information.

Specimens of *Lamellidens corrianus* were collected from fresh water streams near Kariavattom and were immediately used for the investigation. Since it was not possible to obtain urine samples directly, the 'bathing medium' of the animals was used for the estimation of the excretory products[4-8]. Specimens of almost the same size (6 to 7 cm shell length) and weight (15 to 16 g) were selected, their shells cleaned thoroughly and transferred to separate beakers containing 1000 ml of pond water, 24 hours prior to each estimation. 1000 units of pencillin G/ml were added to the water in order to minimise the bacterial action on the excretory products. Five ml portions of the filtered water were used for the estimation of ammonia, urea, uric acid and free amino acids. The ammonia levels were determined by employing the method of Seligson and Seligson[9]. Urea was assayed by conversion to ammonia with urease and later estimating the total ammonia[10]. Uric acid and free amino acids were estimated by following the techniques of Brown[11], and Lee and Takahashi[12] respectively. Blank and control estimations were carried out in all cases.

TABLE I

The pattern of excreation in Lamellidens corrianus. The values denote mean ± Standard Error and represent μg of the substance excreted/total wet weight/24 hours by 15 animals. The numbers in parantheses indicate the % of nitrogen calculated from the total nitrogen of the identified compounds.

Ammonia	Free amino acids	Urea	Uric acid
738·9 ±	152·0 ±	28·2 ±	69·1 ±
SE 22·0	SE 6·9	SE 2·0	SE 3·9
(89·7)	(3·5)	(3·4)	(3·4)

Lamellidens corrianus was found to be predominantly ammonotelic. However, smaller amounts of free amino acids, uric acid and urea were also observed in the excreta.

Although formerly it was considered that the proportion of ammonia nitrogen to the total nitrogen excreted by bivalves is minimal[3], subsequent investigators[4-8,13] have stressed their ammonotelic nature. Thus, Lum and Hammen[8] have recorded the ammonia output in *Modiolus demissus* to be 0·4 to 4·3 mg/100 g total wet weight/24 hours. In *Crassostrea virginica,* of the total 3·4 mg nitrogen excreted/100 g wet weight of tissue/24 hours, ammonia constituted 2·5 mg[6]. Emerson[5] reported that *Mya arenarea* excreted 35 to 125 μg ammonia/g dry weight/hour and *Macoma inconspicua,* 4·9 μg/g dry weight/hour. Investigations by Allen and Garrett[4] revealed that single specimens of *Mya arenarea* eliminated 3·22 mg ammonia/24 hours.

Urea was reported as being excreted by bivalves as early as 1899[14]. It has been shown that urea constitutes 13% of the total nitrogen output in *Crassostrea virginica*[6] and 7% in *Mya arenarea*[13]. In *Lamellidens corrianus* only 3·4% of the total nitrogen of the identified compounds was found to be in the form of urea nitrogen.

The predominance of free amino acids in the excreta of bivalves has attracted considerable attention[15]. Investigations by Lent[13] and Hammen[7] on a number of marine bivalves indicated that the free amino acids constitute the second major excretory product in this group of molluscs. Comparable results have been obtained for *Lamellidens corrianus* also.

There are very few reports on the occurrence of purines in the urine of bivalves. Although Spitzer[3] identified uric acid in the excreta of *Mytilus* and *Unio,* the only recent record on this aspect is that of Hammen[16] on *Crassostrea virginica* in which uric acid amounted to 3% of the total nitrogen output. In *Lamellidens corrianus* 3·4% of the total nitrogen was found to be uric acid.

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---

1. Campbell, J. W. and Bishop, S. H., *Comparative Biochemistry of Nitrogen Metabolism* (Edited by Campbell, J. W.), Academic Press, London, 1970, 1, 103.
2. Florkin, M. and Bricteux Gregoire, S., *Chemical Zoology* (Edited by Florkin, M. and Scheer, T. B.), Academic Press, London, 1972, 7, 301.
3. Spitzer, J. M., *Zool. Jb.,* 1937, 57, 457.
4. Allen, J. A. and Garrett, M. R., *Comp. Biochem. Physiol.,* 1971, 39, 633.

5. Emerson, D. N., *Ibid.*, 1969, 29, 1115.
6. Hammen, C. S., Miller, H. F. and Geer, W. H., *Ibid.*, 1966, 17, 1199.
7. —, *Ibid.*, 1968, 26, 697.
8. Lum, E. C. and Hammen, C. S., *Ibid.*, 1964, 12, 185.
9. Seligson, D. and Seligson, H., *J. Lab. Clin. Med.*, 1951, 38, 324.
10. Conway, E. J., *Microdiffusion Method and Volumetric Error,* Lockwood, London, 1957.
11. Brown, H., *J. Biol. Chem.*, 1945, 158, 601.
12. Lee, Y. P. and Takahashi, T., *Analyt. Biochem.*, 1966, 14, 71.
13. Lent, C. M., *Am. Zool.*, 1969, 9, 283.
14. Cuenot, L., *Archs. Biol.*, Paris, 1899, 16, 49.
15. Potts, W. T. W., *Biol. Rev.*, 1967, 42, 1.
16. Hammen, C. S., *Am. Zool.*, 1969, 9, 309.

A NOTE ON THE SEASONAL CHANGES IN THE INCUBATION TIME OF GROUNDNUT RUST

RUST of groundnut (*Puccinia arachidis* Speg.) is a serious disease[1-4]. The period of incubation reveals the possible disease cycles in a single crop duration. For a proper understanding of the disease, changes in the duration of incubation period are continuously observed for one year, under two different conditions and the results are presented here.

On the 10th of every month, 4 pots, each with 2 plants of 10 days old, were inoculated with uredospore suspension, covered with polythene bags and left in the green house for two days. The bags were then removed and two pots were transferred to a shady region of the green house and the remaining two pots to full sunlight, outside the green house. The plants were observed for the first eruption of pustules.

The incubation period varied from 7 to 18 days under different conditions (Fig. 1). In the shade, the pustules develop one or two days earlier than in bright sunshine. In summer months of May and June incubation period was long, while in winter months (December and January) it was short.

Mc Vey (1965)[5] reported that infection appeared 8 to 10 days after inoculation as whitish flecks, Ramakrishna and Subbayya (1973)[4] reported that the secondary infection readily took place and produced uredosori within 8 days and Bhama (1972)[2] reported that symptoms appeared on the 7th day after inoculation. Present study supports the above observations and further shows that the incubation period changes with the season. That there are seasonal changes in the incubation period of plant diseases is also shown by Jenkyn (1973)[6] who reported seasonal changes in the incubation time of *Erysiphe graminis* f. sp. *hordei* on barley.

Field observations on the incidence of groundnut rust shows that it appears abundantly in July, with the onset of rains, steadily increases upto January and then decreases. From April to June rust incidence is meagre. When the incutation period is short, the rust is widely prevalent and when the period is larger, the incidence of rust is less. Raghunathan and Ramakrishnan (1970)[7], who studied seasonal distribution of rust fungi in Tamil Nadu, also reported a simular trend.

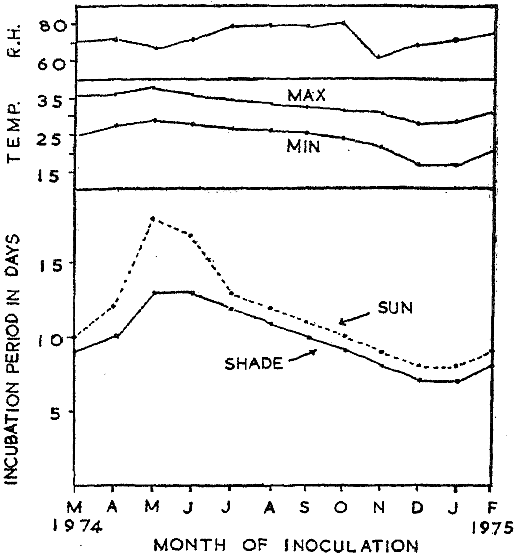

FIG. 1. The temperature and relative humidity recorded at outside the green house area are expressed as monthly means.

I am grateful to late Prof. T. Sreeramulu for guidance. I express my sincere thanks to Prof. A. Seshagiri Rao for encouragement and for going thruugh the manuscript; and to C.S.I.R., for the award of a Research Fellowship.

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1. Jackson, C. R. and Bell, D. K., *Disease of Peanut (Groundnut) caused by Fungi*, University of Georgia, College of Agriculture Experiment Stations, Research Bulletin 56, 1969, p. 47.
2. Bhama, K. S., *Curr. Sci.*, 1972, 41 (5), 188.
3. Sharma, B. D. and Mukherji, S. K, *Ibid.*, 1972, 41 (6), 229.
4. Ramakrishna, V. and Subbayya, J., *Indian Phytopathology*, 1973, 26 (3), 574.
5. Mc Vey, D. V., *Pl. Dis. Reptr*, 1965, 49, 191.
6. Jenkyn, J. F., *Ann. Appl. Biol*, 1973, 73 (1), 15.
7. Raghunathan, A. N.. and Ramakrishnan, K., *Curr. Sci*, 1970, 39, (18), 418.

A NOTE ON THE POLLEN MORPHOLOGY OF *CORCHORUS CAPSULARIS* LINN. *C. OLITORIUS* LINN. AND THEIR HYBRID

THE study of the pollen biology of the Indian jute started with Banerjee[1], and in recent years both pollen morphology and its germination have been studied by several workers[2-5]. The present note relates to the pollen morphology of two cultivated species, namely, *C. olitorius* L. (var. J.R.O. 620) and *C. capsularis* L. (var. J.R.C. 412), and their hybrid with the former as female and the latter as male.

The material for the present study has been supplied by Dr. R. D. Iyer, Division of Genetics, I.A.R.I., New Delhi. The hybrid was running in its ninth generation, and the species *C. capsularis* has been *x*-radiated (20 kr). A notable change in external morphology of the hybrid has been the development of leaf-like stipules, which in both the parents are papillate.

For making pollen preparations, the flower was placed in 70% alcohol, crushed and washed with water and covered with glacial acetic acid followed by acetolysis[3]. The data on pollen morphology are given in Table I.

TABLE I

Data on pollen morphology of two species of Corchorus

Species	Size of pollen grain	Average size of pollen	Equatorial girdle	Size of ora	Shape of ora
C. capsularis Fig. (1 A)	24 μ × 32 μ 28 μ × 25 μ	35 μ × 28 μ	Present	11 μ × 6 μ	slightly lo-longate
C. Olitorius (Fig. (1 B)	39 μ × 32 μ 28 μ × 21 μ	35 μ × 28 μ	Absent	7 μ × 11 μ	la-longate

Pollen grains are 3-zonocolporate in both the species, the endocolpium being slightly lo-longate in *C. capsularis* and la-longate in *C. olitorius.* Further, in *C. capsularis* an equatorial girdle connecting the endocolpium is characteristic, although not well marked, in some grains. The exine is about 3 μ thick and the surface is reticulate.

In the hybrid, *C. capsularis* pollen type (characterised by the lo-longate endocolpium and the equatorial girdle) was seen to dominate forming about 78% as compared to the *C. olitorius* type. It may further be pointed out that the girdle is very firmly developed in the hybrid grains as compared to the one in the parent species, namely, *C. capsularis.* In both *C. capsularis* and *C. olitorius* there are syncolporate and parasyncolporate grains in very minor percentages.

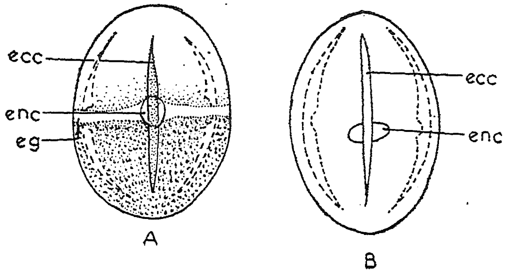

FIG. 1. Palynogram of *Corchorus capsularis* (A) and *C. olitorius* (B). *ecc.*, ectocolpium, *enc.*, endocolpium. Magnification, × 1,000.

Datta[2] described the pollen morphology in the species of *Corchorus* including that of *capsularis* and *olitorius* as tricolpate (rarely tetracolpate and hexacolpate) while Erdtman[3] described the grains in the genus as tricolporate.

In the interspecific hybrid of *C. capsularis* (♂) and *C. olitorius* (♀) the occurrence of *capsularis* pollen type in a high frequency has been quite significant which indicate the dominance of the male character over that of the female. In a similar work, Srivastava *et al.*[5], observed that in the hybrid of *Amaranthus spinosus* × *A. dubius,* the dominance of *spinosus* type grains could be considered to indicate that the species *A. dubius* embodies in it the *A. spinosus* type genome also. In the present study, the *capsularis* grains dominate in the hybrid. However, the *capsularis* type of grains in the hybrid has been found to possess highly developed equatorial girdle as compared to the equivalent parent species, which should be an indication about the genomal changes (mutational or meiotic) undergone by the hybrid, at micro-morphological levels.

The author is grateful to Dr. P. K. K. Nair, for his guidance. She is also thankful to Dr. T. N. Khoshoo, for encouragement and to Dr. R. D. Iyer, for supplying the material.

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1. Banerjee, I., *Indian Jour. Agri. Sci.*, 1933, 3, 116.
2. Datta, R. M., *Phyton*, 1956, 6 (2), 76.
3. Erdtman, G., *Pollen Morphology and Plant Taxonomy, I. Angiosperms*, Stockholm, 1952.
4. Patel, C. I. and Datta, R. M., *Grana Palynol.* 1958, 1 (3), 18.
5. Srivastava, V., Pal, M. and Nair, P. K. K., *Rev. Palaeobot.* and *Palynol.* (In press), 1975.

A MODIFIED SCHEDULE FOR SUGARCANE CHROMOSOMES

Saccharum complex consisting of several species and cultivars poses a formidable problem in cytological investigations due to its high and variable number of small chromosomes and tenacious cell connections. The existing techniques do not yield enough intact cells for reliable chromosome studies besides being too protracted[1,2]. The present study therefore reports a modified Battaglia's fluid[3] and a modified Feulgen stain[4] for effective maceration and rapid processing of root meristems.

The sett roots of hardy cultivars—Co 421, Co 602, Co 740, and Co 1287—are used in the study. The aspects checked are: (*a*) rapidity of fixation, (*b*) maceration, (*c*) nucleo-cytoplasmic contrast and (*d*) preservation of nuclear details. After modification the following fixing fluid, stain and schedule have been found suitable.

Fixing Fluid.—5 parts of absolute alcohol, 2 parts of 2 N HCl, and 1 part each of acetic acid, chloroform, and formaldehyde.

Feulgen Stain.—6 gm more of potassium metabisulphite is added for 100 ml of stain than prescribed by the conventional methods[4].

Schedule.—(1) Fix the roots in the above fixative for 30–45 minutes, (2) wash and store in 70% alcohol at room temperature or proceed directly. Storage at room temperature for a day gave more lasting results in some materials, (3) hydrolyse in N. HCl for 15 minutes, (4) stain in the modified Feulgen stain for 5–20 minutes, and (5) squash in 10% acetic acid.

The present schedule is exceedingly rapid taking only 60–75 min, due to the modified fixative and stain, as against several hours with others[1,2,4] besides ensuing easy and satisfactory maceration into intact cells and preserving cellular contrast.

The authors are thankful to Prof. H. Patnaik, Head of the Department of Botany, Berhampur University and Dr. S. S. Shah, Director, Sugarcane Breeding Institute, Coimbatore.

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Department of Botany, B. N. BEHERA.
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Berhampur, Orissa,
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Coimbatore, *September* 11, 1975.

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1. Jagathesan, D., *Proc. Int. Soc. Sug. Cane Technol.* (Lousiana, 1971), 1972, 14, 303.
2. Rao, P. S., *Sugarcane Breeder's Newsletter*, 1972, 10, 23.
3. Battaglia, E., *Caryologia*, 1957, 9 (2), 368.
4. Darlington, C. D. and LaCour, C. F., *Handling of Chromosomes*, Allen and Unwin, 1969.

DETECTION OF CELL WALL BREAKING ABILITY BY CELLULASE OF *ALTERNARIA BRASSICAE* (BERK.) SACC.

THE cellulolytic enzymes hydrolyse cellulose yielding soluble sugars small enough to pass through the cell walls. From this definition, it follows that these enzymes are extracellular[3]. The edible tissues of higher plants like most fruits, vegetables and roots consist of parenchymatous cells. These are separable into unicells and the naked cell walls are then easily degraded with cellulase[7].

The fungus *A. brassicae* isolated from severe type of leaf spots of mustard (*Brassica campestris* L.) had been observed[2] to produce cellulase constitutively at 5 day incubation period and pH 5·6 at 25 ± 1° C. The addition of 0·5% of L-rhamnose in the basal CMC–N (carboxymethylcellulose-nitrate) medium stimulated the production greatly[1]. The culture filtrate was centrifuged at 5,000 rpm for 10 min. and used as enzyme solution. The ability of this enzyme to degrade the cell walls of potato tuber (*Solanum tuberosum* L.) was investigated. Cylindrical plugs, 8 mm in diameter, were cut from healthy potato tubers and the plugs were injected with distilled water under vacuum for one hour. Discs of 0·4 mm thickness were cut with a sliding hand microtome from these plugs. They were washed quickly with distilled water and stored under it. Five discs were placed in each 3 ml enzyme solution + 2 ml of 0·2 M citrate-phosphate buffer of pH 5·6 in a watch glass. At intervals of 2, 4, 8, 16 and 24 hours, the discs from each watch glass were removed and treated with chlorozinc iodide solution and observed under the microscope for the presence of the cellulosic cell wall. All treatments were in triplicates. The resultant enzyme substrate mixture from each watch glass was tested for enzyme activity by the reducing group method. For the Somogyl's reducing group method[5], 4 ml of the resultant enzyme substrate mixture was taken and assayed at 25 ± 1° C. Likewise, another portion of 4 ml of the mixture was assayed against 1 ml of 1% Na-CMC (sodium-carboxymethylcellulose). The results obtained are presented in the given Table I and Plates A (*i*) and A (*ii*).

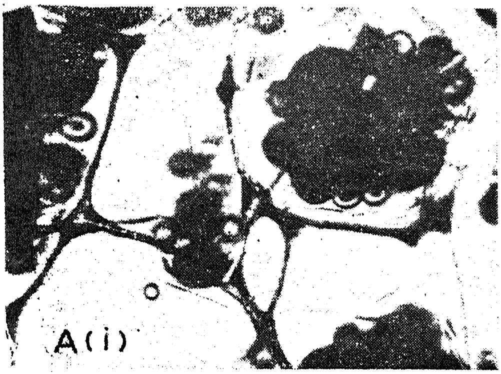

Plate A (*i*)

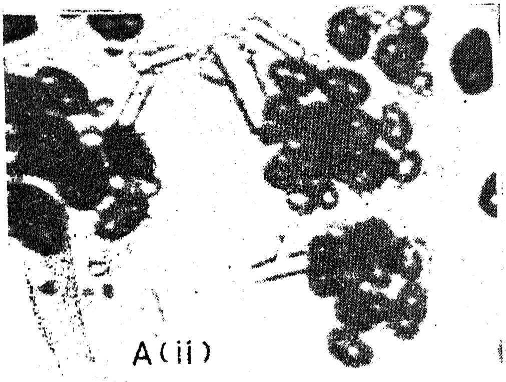

Plate A (*ii*)

It is clear from Table I that cellulase activity was maximum at 24 hours reaction time. This result was further amplified by the histochemical detection as seen in Plates A (*i*) and A (*ii*). When compared with the control Plate A (*i*), the Plate A (*ii*) reveals that, the cell walls are completely broken down and there is no staining by chlorozinc iodide, thus indicating, the degradation of the cellulosic cell walls, by the enzyme cellulase.

The nature of a cell separating enzyme (CSE) was studied by Suzuki *et al.*[6], but the fact that the cellulolytic component of cellulase was responsible for degradation of cell walls of potato was elucidated with *Trichoderma viride* cellulase[4]. Toyama has isolated sweet potato starch using CSE from *Rhizopus*. sp., together with cellulases from *T. viride*[7]. However, the present result is probably the first on the enzymatic degradation of cell walls by only cellulases from *A. brassicae*.

Table I

Reaction time (hrs.)	Cellulase activity at pH 5·6	
	ml $Na_2S_2O_3$	
	Mixture	Mixture + Na-CMC
2	1·3	1·8
4	1·6	2·0
4	2·0	2·5
16	2·7	3·4
24	3·4	4·1

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1. Arun Nehemiah, K. M. and Deshpande, K. B., *Proc. 44th Nat. Acad. Sci. Ind. Absts.*, 1974, p. 56.
2. — and —, *Proc. 62nd Ind. Sci. Cong. Part III, Absts.*, 1975, p. 45.
3. Eriksson, K. E., *Adv. Chem. Ser.* 95. Ed. R. F. Gould, 1969, p. 83.
4. Imai, T. and Kuroda, A., *J. Ferment. Technol.*, 1966, **44**, 854.
5. Somogyi, M. J., *J. Biol. Chem.*, 1945, **160**, 61.
6. Suzuki, H., Abe, T., Urade, M., Nisizawa, K. and Kuroda, A., *J. Ferment. Technol.*, 1967, **45**, 73.
7. Toyama, N., *Adv. Chem. Ser.* 95, Ed. R. F. Gould, 1969, p. 370.

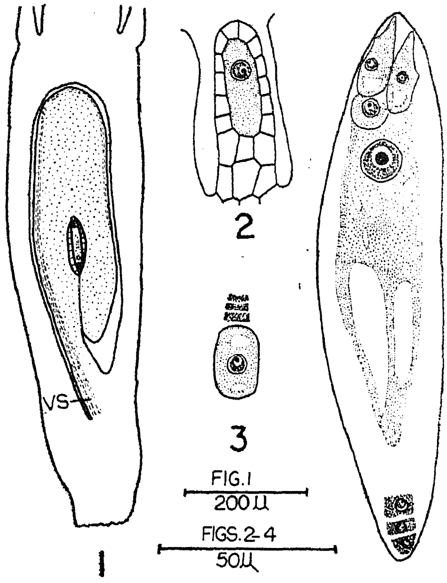
DEVELOPMENT OF FEMALE GAMETOPHYTE IN *HYMENTHERUM TENUIFOLIUM* CASS.

Earlier embryological literature in the family Compositae has been reviewed by Davis[1]. The embryology of *H. tenuifolium,* however, has not been investigated so far and therefore, the present article deals with the development of female gametophyte in this species of ornamental value.

The bicarpellary, syncarpous and unilocular ovary contains a single anatropous, unitegminal and tenuinucellar ovule attached at the base of the locule. The vascular supply of the ovule passes the funiculus and ends on the antiraphe side of the integument (Fig. 1).

The female archesporium is single celled and hypodermal in position. The archesporial cell functions directly as the megaspore mother cell

(Fig. 2). Meiosis in the megaspore mother cell results in the formation of a linear tatrad of megaspores (Fig. 3). The chalazal megaspore is functional and gives rise to an 8-nucleate embryo sac of the Polygonum type as a result of three successive free nuclear divisions. The three antipodal cells are arranged in a linear row and the secondary nucleus lies near the egg apparatus. The synergids are hooked (Fig. 4).



FIGS. 1–4

We are grateful to Dr. T. N. Khoshoo, Dy. Director-Incharge, National Botanic Gardens, for encouragement.

National Botanic Gardens,
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July 14, 1975.

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1. Davis, G. L., *Systematic Embryology of Angiosperms*, John Wiley and Sons, Inc., New York, 1966.

DOTHICHIZA PADI SACC. AND ROUM.— A NEW RECORD TO INDIA

DURING his mycological survey in the hill stations of Maharashtra, the writer collected a fungus belonging to the family Phomaceae on dried twigs of *Calotropis gigantia* R. Br., at Mahabaleshwar. An examination of the fungus revealed the following characters :

Pycnidia subcortical, dark, non-ostiolate, breaking out irregularly ; conidia hyaline, one celled, ovoid to cylindrical.

Based on these observations the fungus was identified as a species of the form-genus *Dothichiza* Lib. The genus was established by Libert in 1880 and the type species is *D. populae* Sacc. and Br. The present fungus was identified as *D. padi* Sacc. and Roum. on the basis of comparative study. This is the first report of its occurrence in India and the description is as follows :

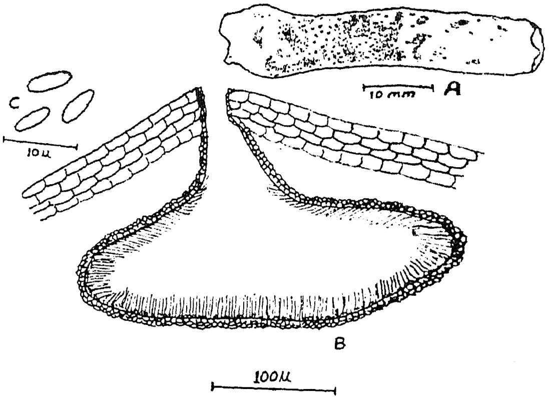

FIG. 1. *Dothichiza padi* Sacc. and Roum. A, Portion of stem showing pycnidia ; B, Pycnidium in section ; C, Conidia.

Dothichiza padi Sacc. and Roum.
Syll. Fung., 1884, 3, 671.

Pycnidia separate, gregarious, erumpent from bark appearing as minute specks on the stems, dehiscing irregularly, subcortical, subglobose to disc-shaped, brown to dark brown, measuring 198·0–409·0 × 49·0–99·0 μ. Conidiophores simple, hyaline, slender, measuring 9·9–16·5 × 1·0–1·7 μ. Conidia hyaline, one celled, ovoid to cylindrical, smooth walled, measuring 6·4–9·9 × 1·7–3·3 μ.

On dry stems of *Calotropis gigantia* R. Br., Mahabaleshwar, 23–12–1974, V. V. Golatkar. Herb. A M H 2565.

The writer is indebted to Dr. P. S. Gharse, Head of the Department of Biology, Ruparel College, Bombay, for his valuable guidance and constant encouragement.

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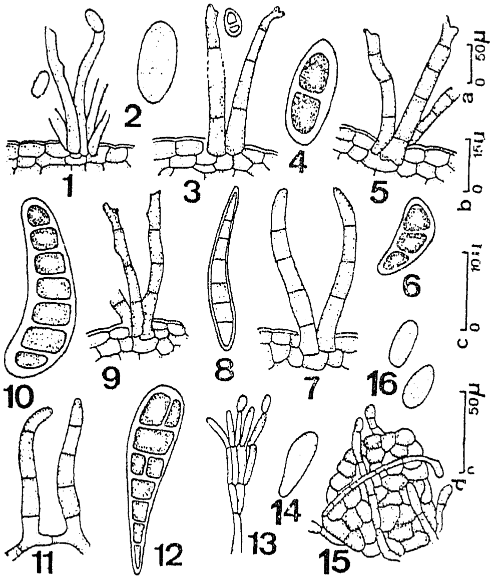
SOME PARASITIC FUNGI ON *CYNODON DACTYLON* FROM VARANASI

Cynodon dactylon Pers., a grass of medicinal value and also used for lawns, is distributed throughout India[2]. A number of pathogenic fungi have been recorded on *C. dactylon*[1,3-5]. Nine parasitic fungi on *C. dactylon* are being reported here for the first time in India, from collections made during the year 1969–70.

1. *Ovularia carneola* Sacc., in *Sacc. Syll. Fung.* 4 : 143. 1886 (Figs. 1 and 2).



Figs. 1–16. Figs. 1, 2. Conidiophores and conidia of *Ovularia carneola.* Figs. 3, 4. Conidiophores and conidia of *Cladosporium herbarum.* Figs. 5, 6. Conidiophores and conidia of *Curvularia lunata.* Figs. 7, 8. Conidiophores and conidia of *Cercospora vernoniae.* Figs. 9, 10. Conidiophores and conidia of *Bipolaris nodulosa.* Figs. 11, 12. Conidiophores and conidia of *Alternaria vitis.* Figs. 13, 14. Conidiophores and conidia of *Myrothecium verrucaria.* Figs. 15, 16. Pycnidium and conidia of *Asteroma martianoffianum.* Scale Line : *a* for Figs. 9 and 13 : *b* for Figs. 2, 4, 6, 14 and 16 ; *c* for Fig. 3 ; *d* for Figs. 1, 5, 7, 8, 10, 11, 12 and 15.

On leaves, near Sankat Mochan Temple, Varanasi. Leg. G. P. Singh, February 17, 1969.

2. *Cladosporium herbarum* Link ex Fries, in *Syst. Mycol.* 3 : 370. 1832 (Figs. 3 and 4).

On leaves, in the ground of Science Faculty, Banaras Hindu University, Varanasi. Leg. G. P. Singh and K. B. Khare, November 25, 1969.

3. *Curvularia lunata* (Wakker) Boedijn, in *Bull. Jard. Bot. Buitenz. Ser.* 3. 13 : 127, 1933 (Figs. 5 and 6).

On leaves, Agriculture Farm, Banaras Hindu University, Varanasi. Leg G. P. Singh and K. B. Khare, October 25, 1970.

4. *Cercospora vernoniae* E. et K., in *Am. Nat.* 1116 : 1883 ; *Sacc. Syll. Fung.* 4 : 431, 1886 (Figs. 7 and 8).

On leaves, Botanical Garden, Banaras Hindu University, Varanasi. Leg. G. P. Singh and K. B. Khare, October 2, 1969.

5. *Bipolaris nodulosa* (Berk. and Curt. ex Sacc.) Shoemaker, in *Can. J. Bot.* 37 : 883, 1959 (Figs. 9 and 10).

On leaves and stems, Old Botanical Garden, Banaras Hindu University, Varanasi. Leg. G. P. Singh, December 30, 1969.

6. *Alternaria vitis* Cavara, in *Sacc. Syll. Fung.* 10 : 679, 1892 (Figs. 11 and 12).

On leaves, Campus of Agriculture Faculty, Banaras Hindu University, Varanasi. Leg. G. P. Singh, September 15, 1969.

7. *Myrothecium verrucaria* (Alb. and Schw.) *Ditm. atud sturm. Deltschl. Cr. Fl.* III, Tab. 4 ; in *Sacc. Syll. Fung.* 4 : 750, 1886 (Figs. 13 and 14).

On leaves, Botanical Garden, Banaras Hindu University, Varanasi. Leg. G. P. Singh, December 27, 1969.

8. *Asteroma martianoffianum* Thum., in *Pilzfl. Sibir. N.* 429 ; in *Sacc. Syll. Fung.* 3 : 202, 1884 (Figs. 15 and 16).

On leaves, Botanical Garden, Banaras Hindu University, Varanasi. Leg. G. P. Singh, February 25, 1969.

9. *Sclerotium rolfsii* Sacc., in *Sacc. Syll. Fung.* 14 : 1139.

On roots, Ayurvedic Garden, Banaras Hindu University, Varanasi. Leg. G. P. Singh, November 10, 1969.

We are grateful to Dr. R. Y. Roy, ex Head, Section of Mycology, Botany Department, Banaras Hindu University, for the help in the identification of the species.

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Varanasi 221 005, *June* 4, 1975.

---

1. Khanna, P. K., *Curr. Sci.*, 1963, 32, 175.
2. Kirtikar, K. R. and Basu, B. D., *Indian Medicinal Plants*, Indian Press, Allahabad, 1918.
3. Payak, M. M., *Indian Phytopath.*, 1949, 2, 190.
4. Roy, R. Y., Dwivedi, R. S. and Khanna, P. K., *Trans. Br. Mycol. Soc.*, 1965, 48, 113.
5. Wehmeyer, L. E., *Mycologia*, 1964, 55, 313.

INDUCED SOMATIC PAIRING IN *NIGELLA SATIVA* L.

PAIRING of homologous chromosomes is generally considered to be a special property of the meiotic cell, with conspicuous exception of *Deptera,* where it apparently occurs in all the cells. Yet, an examination of the cytological literature reveals that the forms in which somatic pairing has been observed include a wide variety of plant and animal species. The discovery that the homologues may undergo pairing in non-meiotic cells was first made in the gonial cells of certain *Hemiptera* by Montgomery[1] (1901) and was reported in the case of spermatogonia of the grass hopper *Brachystola magna* by Sutton (1902).

These observations were extended to somatic cells by Strasburger[2] (1905) who found association of chromosomes in pairs in the embryonic nucleus of the dicot plant *Galtonia candicans* and the monocot *Funkia sieboldiana.*

The authors have studied in detail the effect of mitomycin-C on chromosomes in root tips of *Nigella sativa,* a member of Ranunculaceae, and the phenomenon of Homologous pairing was observed during the course of investigation.

Materials and Methods

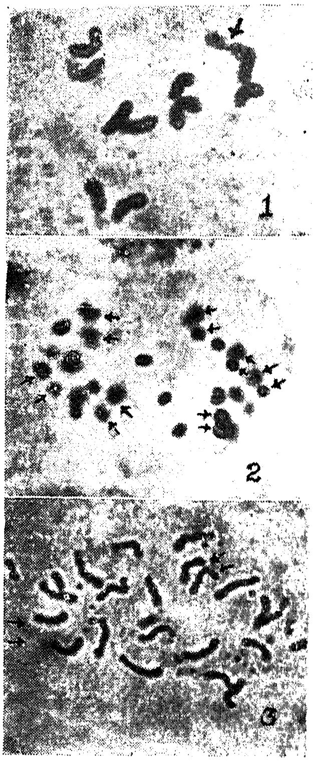
The seeds of *Nigella* were allowed to germinate on moist filter-papers in petridishes at 22° C. After 72 hours the root tips were treated with 0·001% mitomycin-C for intervals ranging from 5 to 40 min. after which the roots were harvested and fixed in 1 : 3 acetic acid alcohol and the cytological preparations[3] were obtained.

The material treated for a period of 5 to 10 min. did not show any observable effect. The cells treated for 15, 20, 30 min. showed increased chromosomal aberrations. After 40 min. the significant chromosomal condensation from prometaphase till anaphase was observed and homologous chromosomal pairing was recorded. In all the other treated material, the chromatid breakages and other abnormalities always appear randomly distributed, so that there is no evidence of any somatic pairing in prometaphase and anaphase chromosomes. Most observations of somatic pairing have been made at metaphase.

In the case of the treated material (15 to 30 minutes) late prophase chromosomes become distinguishable from one another. There is no evidence of somatic association in normal somatic tissue of *Nigella.*

Hence in 40 min. treatment of mitomycin-C (0·001%), only at prometaphase, the homologous chromosomes become juxtaposed to each other with a remarkable regularity (Figs. 1 and 3).

Similarly pairing of homologous chromosom is also clear in anaphase (Fig. 2). In this ca the chromosomes were condensed significantl and were scattered over a very large surface, sma groups of chromosomes remain closely associated a if some force was keeping them together.



FIGS. 1–3. Fig. 1. Arrow shows the telocentric chromosomes are paired with their kinetochores (Telosynapsis) in metaphase. Three pairs of homologues are attached at specific sticky zones and remaining two pairs are juxtaposed, × 1,500. Fig. 2. Arrows show the homologous chromosomes are paired at anaphase. Due to the treatment of antibiotic the chromosomes are condensed, × 1,500. Fig. 3. Arrows indicate the juxtaposition. Some homologues are attached at specific zones in prometaphase, × 1,500.

To explain how homologous chromosome approach each other, we presume that these movements may be due to the kinetochore activity which causes normally congregation of chromosomes towards the equitorial plate of the spindle and which does not occur contemporaneously in all the chromosomes (Colembera[4], 1973). The attached chromosomes seen in (Fig. 1), are telocentric. The end to end chromosome pairing (Telosynapsis) occurs most frequently in homologues. The association shows the importance of kinetochore somatic pairing.

Juxtaposition of homologues is explained by the action of a specific sticky zone located along the chromosome length at corresponding points (Fig. 1). Once the homologues are close to each other they would dispose themselves in the most stable arrangement, preferably where sticky zones are juxtaposed. In normal metaphase and anaphase, this stickiness is not present, but in the material under investigation the stickiness may have developed on the chromosomes due to the effect of mitomycin-C.

In other organisms the somatic pairing is of different patterns. An analogous type of association was reported in plants by Feldman and Mello–Sampayo[5] (1966) where they presume that the kinetochore activity was responsible for the position of homologues near each other. In view of the present observation, we are inclined to believe that *kinetochores are responsible for placing homologues near each other as reported by Feldman and Mello–Sampayo (1966)*. In addition, our observations have support the hypothesis of Wagenaar[6] (1969) where stickiness is attributed to be a factor for association between homologous chromosomes. There is no particular evidence to support the views of Kitani[7] (1963) that the homologous chromosomes are always associated in somatic tissue which is also not supported by workers like Brown and Stack[8] (1968) and Colembera (1973).

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---

1. Montgomery, T. H., *Trans. Am. Phil. Soc.*, 1901, p. 20.
2. Strasburger, E., *Jahrb. Wiss. Botan.*, 1905, 42, 1.
3. LaCour, L., *Aceto-orcien, S.T.*, 1941, 16, 169.
4. Colembera, D., *Caryologia*, 1973, 26 (1), 27.
5. Feldman, M., Mello–Sampayo, T. and Sears, E. R., *Proc. Natl. Acad. Sci. U.S.*, 1966, 56, 1192.
6. Wagenaar, E. B., *Chromosoma*, 1969, 76, 410.
7. Kitani, Y., *Japan J. Genet.*, 1963, 38, 244.
8. Stack, S. M. and Brown, W. V., *Bull. Torr. Bot. Club.*, 1968, 95, 369.

SHORT SCIENTIFIC NOTES

A Waxless Mutant in Rai (*Brassica juncea* Coss)

Very few seemingly different plant types had been observed in the population of an improved variety of yellow *sarson* (*Brassica campestris* var. *sarson*), *viz.*, B–9, at the Pulses and Oilseeds Research Station, Berhampore (W.B.), as early as 1970–71. These types were devoid of any waxy particles on the stem and leaves and appeared to be oily and shiny. Such non-waxy plants were subsequently noticed in brown *sarson* (*B. campestris* var. *dichotoma*), *toria* (*B. campestris* var. *toria*) and *rai* (*B. juncea*). It is seen that such non-waxy mutant was already known in the former three Brassica groups and breeding studies revealed it to be a monogenic recessive character[1]. Srinivasachar and Malik[2] reported the waxless mutant in the irradiated material of *B. rapa*. Thompson[3] claimed such mutant in *B. oleracea* var. *acephala*. They also indicated the association of the non-waxy character with aphid resistance. The occurrence of the non-waxy mutant in the *juncea* group of Brassica is being reported here from India, for the first time. The waxless mutants of *rai* and yellow *sarson* in the present observation showed that the plants were less damaged by aphid than their waxy counterparts. Further work on the utilization of the mutants of these varieties is in progress.

The authors are grateful to Dr. D. Mukherjee, Joint Director of Agriculture (Research) and Dr. K. Sengupta, Plant Breeder (Oilseeds), Government of West Bengal, for their keen interest in the work. Thanks are also due to the ICAR, and the Directorate of Agriculture, West Bengal, for providing facilities.

Pulses and Oilseeds D. K. NATH.
Research Station, S. MUKHOPADHYAY.
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Revised MS on *October* 30, 1975.

---

1. Singh, D., *Rape and Mustard,* ICOC, Hyderabad, 1st ed., 1958, p. 12.
2. Srinivasachar, D. and Verma, P. K., *Curr. Sci.*, 1971, 40, 311.
3. Thompson, K. F., *Nature, Lond.*, 1963, 198, 209.

Metarrhizium anisopliae (Metch.) Sorokin on Brinjal Mealy Bug and its Use in the Biological Control of the Pest

There are many hyphomycetous fungi which grow on insects. The most common parasitic species belong to the genera *Beauvaria, Metarrhizium, Isaria* and *Spicaria*[1]. Among these the genus *Metarrhizium* is strictly entomogenous[2]. *Metarrhizium anisopliae* (Metch.) Sorokin, the cause of 'green muscardine' disease of diverse insects is the commonest species of *Metarrhizium*. In India its occurrence has been reported from a few insect species[3]. Narasimhan (1970)[2] held that the species has a potential importance in the biological control of insect pests. This note records *M. anisopliae* on *Centrococcus insolitus* Green, the brinjal mealy bug. Brief description of the species and observations on biological control of the pest are also given.

Mealy bug colonies on brinjal plants in the fields of Nadia District, West Bengal, were seen to be infected by the fungus, from November to March when the temperature ranged from 10–25° C and the maximum relative humidity remained lower than 75%. The infected colonies, particularly the ovisacs, instead of being cottony white, appear dark bluish green and powdery, due to sporulation of the fungus at the surface.

The fungus was brought into pure culture in honey-peptone agar (HPA) from spore mass on the infected insects. It could also be isolated from surface-sterilized eggs, indicating that, in the nature, eggs are also infected. Colonies on HPA and Sabourad's maltose agar start as white, fluffy mycelial growth which soon change to green from the center and ultimately become dark olive green. Sporophores and hyphae in culture extend upwards, being closely packed, and the branches remain intertwined. Conidia formation takes place by abstriction of the elongated buds which appear at the distal end of sporophores branching in the tip in the manner of a fork. Budding continues basipetally, forming chains of conidia. Chains of conidia arising from adjacent sporophores cohesce as prismatic masses. In old cultures they form a crust over the surface. Conidia are at first white and later become olive green. Conidia from culture measure 5·5–7·9 μ × 2·3–3·5 μ, the range of length being higher on insects (3·5–8·5 μ). Pathogenicity of the fungus to brinjal mealy bug could be proved by inoculation of the healthy insects which were grown from eggs on potato sprouts in sterile glass containers, and 2nd–3rd instars were dusted with spore mass. Seven to ten days after inoculation the insects were dead. Sporulation was obtained after another three days. On dissecting the dead insects, hyphae were seen to ramify deep in the tissue and conidiophores emerged at the surface. On the basis of the salient characters, the fungus was identified as *Metarrhizium anisopliae* (Metch.) Sorokin, and *Centrococcus insolitus* Green (Pseudococcidae, Homopetra) is a new host record.

Brinjal plants severely infested with the mealy bug in the fields were dusted with spore mass obtained from cultures grown on molasses-peptone agar (5% molasses, 1% peptone) at two seasons, November and February. Within 12–15 days after inoculation, mealy bug colonies on the leaves started dying and plants gained vigour after about a month. Approximately 80 and 56% of the inoculation tests were successful at the respective seasons. Judging by the results of inoculation tests and the performance of plants after inoculation, it appears that mealy bug infestation of brinjal can successfully be eradicated by using *M. anisopliae*.

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1. Madelin, M. F., *Diseases Caused by Hyphomycetous Fungi*, In : *Insect Pathol*, E. A. Steinhaus, ed., Academic Press, New York, 1963, 2, 233.
2. Narasimhan, M. J., *Ind. Phytopathol.*, 1970, 23, 16.
3. Ram Raje Urs, N. V. and Govindu, H. C., *Mycopath. Mycolog. et Appl.*, 1971, 44, 317.

Natural Control of Ground-nut Aphid, *Aphis craccivora* Koch. in Central Gujarat

The aphid, *Aphis craccivora* Koch. is a very serious pest of ground-nut, *Arachis hypogaea* L. throughout the State. During August, 1974 severe infestation by this species was noticed in ground-nut crop in Kapadwanj taluka. The pest was first observed in the second week of August when 10% of the plants were found infested. The aphid infestation spread very rapidly and built up large colonies on the tender shoots of different branches to the extent of 97% infestation during the third week of August.

The predators collected at this time revealed two species of coccinellids, *Menochilus sexmaculatus* (F.) and *Coccinella septempunctata* L.; one syrphid, *Xanthogramma scutellare* (F.) and *Chrysopa carnea* Steph. The aphid population decreased with the increased population of the natural enemies and by the fourth week of August *M. sexmaculatus* was quite abundant to bring about complete control of the aphid. The counts of predators made during the week in 5 different fields showed that each plant had on an average 5 grubs, 20 pupae and 10 adults of *M. sexmaculatus.* The population of *C. septempunctata* and *X. scutellare* was very low to record only 0·6 and 0·4 pupae per plant respectively. The incidence of *Chrysopa carnea* was negligible. The population of the predators declined by the first week of September when each plant had only 6 adults of *M. sexmaculatus* indicating that most of them must have migrated to other areas due to absence of food in the crop.

Thus, *M. sexmaculatus* played a very important role in controlling such a severe infestation of aphid as a natural control agent saving the cost of at least one round of insecticidal application. This also indicates that it is possible to check the aphid infestation in other endemic areas by early releasing of laboratory reared *M. sexmaculatus* adults.

The authors are indebted to Dr. R. M. Patel, Director of Campus for the facilities and interest in the work.

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Anand-Campus, Anand,
August 5, 1975.

Residues of Certain Systemic Insecticides in Sweet Potato Tubers

Sweet potato weevil (*Cylas formicarius*) is controlled by spraying 0·1% fenthion or carbaryl and the residues of these pesticides in the sweet potato tubers were found below the F. D. A., U.S.A. tolerance level (Subramaniam *et al.,* 1974). In this investigation the soil application of certain systemics Temik, Carbofuran and Disyston granules at 1·00 kg a.i/ha were made at the time of tuber formation for the control of Sweet potato weevil. The experiment was laid out in a randomised block design with three replications. The fresh tuber samples collected at the time of harvest were analysed for the residues of Temik (Jhonson and Stansburg, 1966), Carbofuras (Gupta and Dewan, 1971) and Disyston (Schumann and Olson, 1964).

The results of analysis are as follows :

Insecticide	Residues in ppm	FDA tolerance level (ppm)
Temik	0·095	0·20
Carbofuran	0·105	0·20
Disyston	0·326	0·75

The study indicated that all the three insecticides left residues in the tuber well below the tolerance level of FDA of USA indicating that they can be safely applied in the soil at 1·0 kg a.i/ha to control the sweet potato weevil.

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1. Gupta, R. C. and Dewan, R. S., *All India Symp. on Prog. and Problems in Pesticide Residue Analysis,* Ludhiana, 1971, p. 208.
2. Jhonson, D. P. and Stansburg, H. A., *J. Assoc, Off. Anal. Chem.,* 1966, 49, 399.
3. Schumann, S. and Olson, T., *Chem. Agro Corporation, Publication No. 8544,* 1964, p. 1.
4. Subramaniam, T. R., Thirumurthi, S., Asaf Ali, K., Thangavel, P. and Shanmugam, A., *South Indian Hort.,* 1974, 22, 137.

Cuscuta reflexa—A New Host for *Aphis craccivora*

While rearing *Aphis craccivora* (a virus vector) and *Cuscuta reflexa* (virus carrier) on different citrus plants, to study their efficiency in transmitting Tristeza virus of citrus, a large number of aphids migrated to *C. reflexa* and started feeding and breeding on the tips. Laboratory studies revealed that a single adult produced 10–12 nymphs in life span of 14–20 days and each nymph became adult in 6–7 days.

A. crassivora has already been reported to feed on *Dolichous lablab, Cassia tora,* sweet orange, grape fruit and Rangpur lime (Verma *et al.,* 1965). *Cuscuta reflexa* thus appears to be a new host for the aphid.

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Ludhiana, *September* 18, 1975. S. K. KAPUR.

1. Verma, P. M., Rao, D. G. and Capoor, S. P., "Transmission of Tristeza Virus by *Aphis craccivora* Koch and *Dactynotus jaceae,*" *Indian J. Ent.,* 1965, 27, 67.

Production of Teliospores in Artificial Culture of *Ustilago crameri* (Kornicke)

Ustilago crameri the smut disease on *Setaria italica* (Linn). Beauv. is ubiquitous in its distribution and cause heavy damage and loss in several States in our country particularly in South India. In severe case of infection, 75% of the grains in the ears become smutted though rarely the terminal portion of the spike may escape. The sori are 2–4 mm in diameter and appear slightly larger than the grains.

The spores are round or angular in shape, and dark-brown in colour. They are smooth and measure 7–10 μ. Germination takes place without any rest period. During spore germination studies, it was observed that culture maintained by mono-teliospore isolations develop teliospores in potato-sucrose peptone malt agar after a period of six weeks at 25° C. The teliospores formed in culture were more or less spherical, smooth and often slightly larger than the teliospores produced in the host plant. Similar type of teliospores formed in culture are reported earlier by Govindu and Fischer[1], for *Ustilago striiformis* (West.) Niessl.

The present report is the first record for the occurrence of teliospores in culture of *Ustilago crameri*. Germination studies of the teliospores formed in artificial culture as well as their method of formation and electron microscopy of morphology of surface structure are under investigation.

Department of Plant Pathology, — B. Vijayaraghavan.
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1. Govindu, H. C. and Fischer, G. W., "Physiological variation in the stripe smut of grasses, *Ustilago striiformis*," Research Studies, Washington State University, Pullman, U.S.A., 1962, **30** (4), 139.

The Natural Occurrence of *Bipolaris tetramera* on Termites

In October, 1974 at Varanasi, the authors observed stored woollen cloth being destroyed by a group of termites (*Odontotermes obesus* Rambur) with fungal infection.

This fungus was isolated on PDA and identified as *Bipolaris tetramera* (Mc Kinney) Shoemaker. Pathogenicity tests were conducted on living, healthy termites already kept under isolation for 15 days to ensure the absence of any other microorganism. The healthy termites were placed in sterile Petri-dishes lined with filter-papers, and inoculated with spore-*cum*-mycelial suspension of *Bipolaris tetramera*. All termites inoculated with fungal suspension died within 72 hours, while control individuals treated with sterile distilled water remained alive and healthy up to 15 days. Re-isolations from the artificially infected termites revealed the same fungus.

There appears to be no previous record of any species of *Bipolaris* on termites. *B. tetramera* though commonly a soil saprophyte has also been reported to be a weak pathogen on various economic plants. In view of this, the present report would indicate the possibility of biological control of termites.

Grateful thanks are due to Dr. R. Y. Roy, for his keen interest in the present work and one of us (KBK) also wishes to acknowledge indebtedness to the C.S.I.R., for appointment in the Scientist's Pool.

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1. Subramanian, C. V., *Hyphomycetes*, I.C.A.R., New Delhi, 1970, p. 758.

A Short Note on *Frerea indica* Dalz.

Frerea Dalz. a monotypic genus, is represented by the endemic species *F. indica* Dalz., confined to the Sahyadri ranges of Maharashtra. Except for a solitary collection in June 1956 (Puri 2619, BSI) the plant has not been collected from its type locality at Junnar, since its original collection by Dalzell in 1864 and subsequently by McCann (McCann 137/18 H BLATT) though it has been recently reported from Kate's point at Mahabaleshwar and Purandhar fort as well. On the basis of the succulent stems and rotate corolla, Rowley (*Nat. Cact. and Succ. J.* **30** : 78, 1958) merged it under *Caralluma* R. Br. renaming it as *C. frerei* Rowley, but the profuse leafy nature, distinct floral features and the absence of any intermediate species of *Caralluma* with the fleshy large leaves justify the retention of *Frerea* as a distinct genus.

Somatic squashes after pre-fixation with 8-oxyquinoline for 2 hours and staining in acetic-orcein following the normal procedures revealed $2n = 44$ (voucher specimen : *Singh* 108937, BSI) which is a first report for the species. Though Dalzell has described the follicles and seed, the species has not normally been found to fruit in nature but under experimental cultivation, the plants fruited for the first time at Pune in 1971, the seeds exhibiting 50% germination. A redeeming feature of this rare plant is its successful ready propagation from cuttings and it is worth introducting in garden as an ornamental pot plant.

Botanical Survey of India, — R. Sundara Raghavan.
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Poona-1, *September* 10, 1975.

A New Species of Septoria on Hops (*Humulus* Spp.) in Kashmir

Hops is grown in Kashmir valley by the Department of Agriculture and other agencies for many years. Its flowers are used by various breweries in India and lot of foreign exchange is thus saved by making use of them.

During our survey of Hops (*Humulus* Spp.) plantation at the Agricultural Experimental Research Station, Shalimar, a new disease has been found manifesting itself on its leaves, particularly on local and late cluster varieties. The disease appears as small necrotic spots, which later on coalesce and forms bigger patches. Initial infection appears to resemble that of leaf necrosis of virus origin. The spots turn brownish or rusty with age.

The pathogen responsible for the disease has been identified as *Septoria lupulina* Ellis and Kellerman (IMI 194898). Cluster of pycnidia appear in the centre of the spot, which can also be traced at its periphery; pycnidia which measure 140-180 µ are found with small ostiole. Conidia are slender, faintly septate with slightly curved ends. They measure 30-48 × 2-2.5 µ.

The species *Septoria lupulina* on hops is a new record for India. The pathogen was isolated on PDA from diseased leaves. The fungus grew well and a number of pycnidia bearing cluster of conidia were observed.

The disease incidence was estimated at 10-15% in most of the areas, which was even higher in other fields.

The authors express their gratitude to Dr M. B. Ellis, Commonwealth Mycological Institute, Kew, Surry (England), for confirming the species.

Grateful thanks are also due to Mr A. G. Malik, Director of Agriculture, J & K Government, for the facilities and his keen interest in this study.

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Zinnia elegans Jacq. The Latent Host of Watermelon Mosaic Virus

The watermelon mosaic virus (WMV) was previously known to have its host range limited to the family Cucurbitaceae only[1]. Later studies by Freitag[2] and Grogan[3] suggested that watermelon mosaic virus can infect the plants of other families also including Leguminoceae, Malvaceae and Umbelliferae.

On the basis of host range studies Webb and Scott[4] divided watermelon mosaic virus complex into two groups: WMV 1 limited to Cucurbitaceae only and WMV 2 infecting plants of other families also. Milne and Grogan[5] indicated them to be the different strains of the same virus.

In the present study, the virus isolate was obtained from naturally infected plants of *Trichosanthes dioica* Linn. The host range studies were carried out with twenty-three plants belonging to eleven different families. The virus was recovered by back inoculation from *Zinnia elegans* Jacq. only. The virus was recovered by insect transmission also using *Aphis gossypii* as vector. Detailed studies made on the physical properties, insect transmission, particle morphology and symptoms induced in cucurbits indicated it to be a strain of watermelon mosaic virus, Bhargava and Tewari[6].

This report is the first record of this plant as a symptomless carrier for watermelon mosaic virus and hence an indication of the important host for perpetuation of the virus.

Grateful thanks are due to Prof. K. S. Bhargava, Head, Department of Botany, Gorakhpur University, for guidance and to CSIR, New Delhi, for financial assistance.

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---

1. Anderson, C. W., *Phytopathology*, 1944, **44**, 190.
2. Freitag, J. H., *Ibid.*, 1952, **42**, 8 (Abstr.).
3. Grogan, R. G., Hall, D. H. and Kimble, K. A., *Ibid.*, 1959, **49**, 366.
4. Webb, R. E. and Scott, H. A., *Ibid.*, 1965, **55**, 895.
5. Milne, K. S. and Grogan, R. G., *Ibid.*, 1969, **59**, 809.
6. Bhargava, K. S. and Tewari, J. P., *Plant. Dis. Reptr.*, 1970, **54** (8), 727.

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---

1. Govindu, H. C. and Fischer, G. W., "Physiological variation in the stripe smut of grasses, *Ustilago striiformis*," Research Studies, Washington State University, Pullman, U.S.A., 1962, 30 (4), 139.

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1. Subramanian, C. V., *Hyphomycetes*, I.C.A.R., New Delhi, 1970, p. 758.

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A New Species of Septoria on Hops (*Humulus* Spp.) in Kashmir

Hops is grown in Kashmir valley by the Department of Agriculture and other agencies for many years. Its flowers are used by various breweries in India and lot of foreign exchange is thus saved by making use of them.

During our survey of Hops (*Humulus* Spp.) plantation at the Agricultural Experimental Research Station, Shalimar, a new disease has been found manifesting itself on its leaves, particularly on local and late cluster varieties. The disease appears as small necrotic spots, which later on coalesce and forms bigger patches. Initial infection appears to resemble that of leaf necrosis of virus origin. The spots turn brownish or rusty with age.

The pathogen responsible for the disease has been identified as *Septoria lupulina* Ellis and Kellerman-(IMI 194898). Cluster of pycnidia appear in the centre of the spot, which can also be traced at its periphery pycnidia which measure 140–150 μ are round with small ostiole. Conidia are slender, faintly septate with slightly curved ends. They measure 30–45 × 2–2·5 μ.

The species *Septoria lupulina* on hops is a new record for India. The pathogen was isolated on PDA from diseased leaves. The fungus grew well and a number of pycnidia bearing cluster of conidia were observed.

The disease incidence was estimated at 10–15% in most of the areas, which was even higher in other fields.

The authors express their gratitude to Dr. M. B. Ellis, Commonwealth Mycological Institute, Kew, Surry (England), for confirming the species.

Grateful thanks are also due to Mr. A. G. Malik, Director of Agriculture, J & K Government, for the facilities and his keen interest in this study.

Mycologist, Agriculture Research Station,
Shalimar (Kashmir),
and
Research Assistants,
Mycology and Plant Pathology Section,
Shalimar (Kashmir),
July 16, 1975.

RAJ KUMAR MIRAKHUR.
SHIV JEE DHAR.
ASHOK KUMAR DHAR.

Zinnia elegans Jacq.—The Latent Host of Watermelon Mosaic Virus

The watermelon mosaic virus (WMV) was previously known to have its host range limited to the family Cucurbitaceae only[1]. Later studies by Freitag[2] and Grogan[3] suggested that watermelon mosaic virus can infect the plants of other families also including Leguminoceae, Malvaceae and Umbelliferae.

On the basis of host range studies Webb and Scott[4] divided watermelon mosaic virus complex into two groups: WMV–1 limited to Cucurbitaceae only and WMV–2 infecting plants of other families also. Milne and Grogan[5] indicated them to be the different strains of the same virus.

In the present study, the virus isolate was obtained from naturally infected plants of *Trichosanthes dioica* Linn. The host range studies were carried out with twenty-three plants belonging to eleven different families. The virus was recovered by back inoculation from *Zinnia elegans* Jacq. only. The virus was recovered by insect transmission also using *Aphis gossypii* as vector. Detailed studies made on the physical properties, insect transmission, particle morphology and symptoms induced in cucurbits indicated it to be a strain of watermelon mosaic virus, Bhargava and Tewari[6].

This report is the first record in this plant as a symptomless carrier for watermelon mosaic virus and hence an indication of the important host for perpetuation of the virus.

Grateful thanks are due to Prof. K. S. Bhargava, Head, Department of Botany, Gorakhpur University, for guidance and to CSIR, New Delhi, for financial assistance.

Department of Botany,
M.L.K. College,
Balrampur (U.P.), *September* 1, 1975.

J. P. TEWARI.

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1. Anderson, C. W., *Phytopathology*, 1944, 44, 190.
2. Freitag, J. H., *Ibid.*, 1952, 42, 8. (Abstr.).
3. Grogan, R. G., Hall, D. H. and Kimble, K. A., *Ibid.*, 1959, 49, 366.
4. Webb, R. E. and Scott, H. A., *Ibid.*, 1965, 55, 895.
5. Milne, K. S. and Grogan, R. G., *Ibid.*, 1969, 59, 809.
6. Bhargava, K. S. and Tewari, J. P., *Plant. Dis. Reptr.*, 1970, 54 (8), 727.

REVIEWS AND NOTICES OF BOOKS

High Energy Lasers and Their Applications—Physics of Quantum Electronics (Vol. 1). S. F. Jacobs, M. Sargent III, and M. O. Scully (Editors). (Addison-Wesley, U.S.A.), 1974, Pp. x + 411. Price $ 17.50.

This book is based on lectures delivered by well known scientists during the 1973 Summer School on the Physics of Quantum Electronics sponsored by the University of Arizona. The first part of the book contains contributions on laser physics. The second section is a review of some aspects of laser fusion, which, the title notwithstanding, is the only laser application discussed.

In Chapter I (46 pp.), M. Sargent III presents a survey (unfortunately marred by several typographical errors) of one- and two-mode strong signal laser theory, using both the rate equation and the semiclassical approach. The plausibility arguments and numerical illustrations that replace detailed derivations usefully complement the more formal papers of Lamb and co-workers. H. Shih, M. O. Scully and S. F. Jacobs (Chapter II, 29 pp.) describe an ingenius new model of the unstable resonator that enables a generalised and integrated analysis to be made of the effects of the active medium, non-linear mode coupling and diffraction, and the mode behaviour to be continuously predicted through the stable-unstable transition. F. A. Hopf (Chapter III, 100 pp.) reviews laser amplifier theory for homogeneously, inhomogeneously and power broadened media. Illustrative examples are drawn from actual systems, but the section on multiline molecular laser amplifiers is now slightly outdated. This masterly survey contains a great deal of information not available in scientific journals. In Chapter IV (70 pp.), C. B. Hogge considers the effects of turbulence and thermal blooming on laser beams propagating through the atmosphere, with supporting experimental results and computer simulations of the effects of thermal blooming on the density profile. The laser physics section concludes with Chapter V (46 pp.) by P. V. Avizonis on the inversion and relaxation processes occurring in electrical and gas dynamic CO_2 lasers and the chemical HF laser. The surprising neglect of the fundamental Treanor, Bray, and Sharma-Brau theories of vibrational relaxation, and the electron distribution calculations of Nighan weakens an otherwise adequate discussion.

The section on laser induced fusion (LIF) begins with a lucid semi-qualitative description by K. Beyer (Chapter VI, 40 pp.) of the physics of initiating thermonuclear fusion by light absorption, the capabilities of available Nd : Glass and CO_2 lasers, and the design constraints of LIF reactors. J. R. Murray and P. W. Hoff (Chapter VIII, 58 pp.) compare the characteristics of the hypothetical laser ideal for LIF with both presently available as well as futuristic lasers. A calculation by R. Morse (Chapter VIII, 16 pp.) to estimate the magnitudes of quantities important in LIF concludes the book.

By presenting recent developments (some of the material is just beginning to appear in scientific journals) in the field of lasers in an insightful and informal style, this book bridges the gap between the text book and the scientific journal. It should prove useful to workers in quantum electronics wishing to keep abreast of areas outside their own specialisation.

Since most of the discussions start from first principles, this book should also provide students interested in the field, with a sound base from which to embark into the literature.

J. P. Pichamuthu.

Pattern Recognition Principles. By J. T. Tou and R. C. Gonzalez. (Addison Wesley, Reading, Mass., U.S.A.), 1974. Pp. xi + 377. Price U.S. $ 19.50.

In 1965, there were only two books that could be considered as text books for pattern recognition (PR)—Nilsson's 1965 monograph and Sebestyen's 1962 work. Today there is a score of books to choose from. (*See* T. M. Cover, *IEEE Trans. IT*, Nov. 1973 for an excellent review of the previous books.) This new book is a welcome addition to this set.

The book is divided into eight chapters. Chapter 1 introduces the reader to the problem *via* the information explosion of twentieth century and the evolution of a methodology to cope with the enormous data arising in a large number of situations. Examples from OCR, remote sensing, finger print classification and nuclear reactor surveillance provide excellent motivation for the student. Chapter 2 and Chapter 3 deal with decision functions, distance functions and some clustering techniques and their applications to the pattern classification. Pattern classification by likelihood functions and Bayes techniques are considered in Chapter 4. Chapters 5 and 6 deal with trainable pattern classifiers, covering gradient algorithms, potential functions and stochastic approximation. Chapter 7 considers the important problem of pattern preprocessing and feature selection. Chapter 8 introduces the new approach based on Syntactic Pattern Recognition and is interesting.

Thus, the book introduces a large number of techniques in a simple manner with neat diagrams and explanations. The book is particularly suitablt for a graduate course in view of the problems provided at the end of each chapter and adequate coverage of a number of useful topics.

However, some of the important concepts such as results concerning dimensionality, sample size and error estimation, fuzzy sets are not considered. Also a particular application should have been highlighted althrough like the scene analysis in Duda and Hart's (Wiley, 1973) book.

B. L. DEEKSHATULU.

Biological Transport. By H. N. Christensen. (Second Edition, Revised, Enlarged and Reset), Publishers : W. A. Benjamin, Inc., Reading, Mass., U.S.A.

Living cells have developed membranes to separate two phases. Implicit in this design is the transport of materials across. Membranes have become barriers and carriers at the same time and intrinsically selective. Transport phenomenon is universal and concerns all biological materials from simple protons to complex macromolecules, all aspects of life processes from intestinal absorption to nerve conduction, and all branches of biosciences from phenomenological description to molecular characterization. This is one field wherein botanists, zoologists, geneticists, microbiologists, nutritionists, physiologists, biochemists, neurochemists and biophysicists have all contributed, and, as claimed by the author, caused "fragmentation of the subject among the disciplines with particular emphasis on molecular approaches". This book true to the claim has put the whole subject together and synthesized the common features of transport in biological systems. It has surveyed the scope of the problem of transport, described the architecture of the membranes and discussed the thermodynamics and kinetics of transport processes. It has considered the transport systems of different solutes, the approaches from genetic studies and the specific binding proteins. Group translocation which is a form of transport with the distinction of alteration of chemical form—vectorialization of a chemical reaction—has been aptly included. The multiple ATPases in membranes have been described and an attempt is made to understand their participation in the energy transactions incidental to transport. A full chapter is devoted to transport in mitochondria. The incisive discussion on the techniques in transport studies and some procedures at the end of the book are likely to prove the most useful for those who wish to embark on the experimental work. The terms such as "transport", "active transport", "permease" and "efflux" have been subjected to semantics and the author attempts to give guidelines for terminological clarity. The book has a personalized style as though a teacher is talking to students in a class-room—asking question and giving answers and thus giving away the subject matter. All through, the chapters are characterized by an "introductory" statement which projects the basis of the subject, and by a "concluding" or "summary" comment which not only is an epitome of the essay but posed the problems yet unsolved. In fact one gets a glimpse of the whole book by reading these two items. This is a commendable new style. For an "Advanced Book Program" from an international publishers, W. A. Benjamin, Inc., the quality of production should have been better. For instance, the figures in some places are out of proportion and the reproduction of the lines poor, and the indexing of the pages (360–406 and 442–452) was wrong in step by 1–2 pages, which causes a momentary irritation to the reader. Notwithstanding this, the book serves as an excellent companion for a teacher giving a course or a research workers as a reference source.

T. RAMASARMA.

5560/75. Published by S. R. S. Sastry, Manager, Current Science Association, Bangalore-560006
and Printed at The Bangalore Press, Bangalore-560018.

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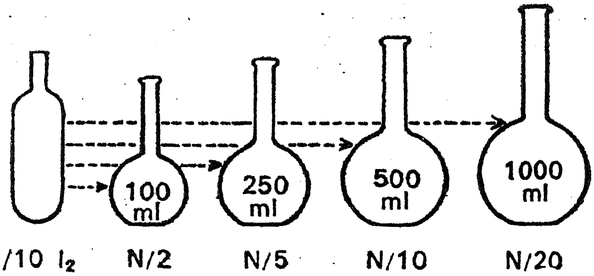

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„ „	N/5	„	„	„	„	250 ml
„ „	N/2	„	„	„	„	100 ml

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DIFFRACTION IMAGES OF TRUNCATED PERIODIC OBJECTS IN PANORAMIC CAMERAS IN THE PRESENCE OF PARABOLIC IMAGE MOTION

A. K. GUPTA AND K. SINGH*

Department of Physics, Indian Institute of Technology, New Delhi 110029

ABSTRACT

Irradiance distribution in the diffraction images of truncated one-dimensional periodic targets formed in the presence of parabolic image motion is discussed. Incoherent objects with sine and square wave profiles are considered in the case of an optical system with circular aperture. Image contrast, tabulated for a number of cases, is expected to provide a valuable tool for an image analyst.

INTRODUCTION

Use of optical transfer function techniques in image evaluation is now very well known and an excellent review of the present status of the subject has been given by Norton[1]. Truncated periodic targets are also useful in many practical situations, when these occur in image format in connection with the detection and resolution in aerial photographs of a wide variety of objects. Consequently the diffraction images of truncated targets have been investigated extensively[2-5].

Despite the best image motion compensation (IMC) techniques, some residual motion is always present[6-8] and this impairs the quality of aerial photographs. Effects of linear and sinusoidal motion on the images of various objects has been extensively investigated[5-7] in the past. Residual image motion in transverse scanning panoramic cameras[7-10] results from the finite slit width which precludes film's IMC from being exactly correct for all images simultaneously exposed in the slit. The image displacement is a parabolic function of time[7,10,12]. Such motion has recently been considered with respect to its effect on imagery of infinite periodic objects[13], holographic measurements[14] and image restoration[15,16].

The present paper discusses some aspects of diffraction images of truncated objects in the presence of parabolic image motion.

2. THEORY

The frequency spectra $I(\omega)$ of the image and $O(\omega)$ of the object are related by

$$I(\omega) = O(\omega) \cdot T(\omega) \tag{1}$$

where $T(\omega)$ is the system transfer function and ω the normalized spatial frequency. The intensity distribution $i(p)$ in the image is then given by

$$i(p) = \int_{-\infty}^{\infty} T(\omega)\, O(\omega)\, \exp(ip\omega)\, d\omega \tag{2}$$

The object intensity function for N-cycle bar and sinusoidal targets are given respectively[2,4], by

$$O(\omega) = (2\pi\omega)^{-1}\,[\sin(2N\omega L)/\cos(\omega L)] \tag{3}$$

$$O(\omega) = \pi^{2}\omega \,.\, \sin(2N\omega L)/(\pi^{2} - 4\omega^{2}L^{2}) \tag{4}$$

where $4L$ is the spatial period. Both p and L are in reduced units.

The overall optical transfer function $T(\omega)$ of the optical system under consideration is the product of incoherent transfer function $C'(\omega)$ of the diffraction limited system and the transfer function $t(\omega)$ for the image motion, *i.e.*

$$T(\omega) = C'(\omega) \,.\, t(\omega) \tag{5}$$

$C'(\omega)$ for the diffraction limited circular aperture is given by

$$C'(\omega) = 2/\pi\,[\cos^{-1}(\omega/2) - \omega/2\,(1 - \omega^{2}/4)^{\frac{1}{2}}] \tag{6}$$

The OTF, for parabolic image motion is given[7,10,11] by

$$t(\omega) = \frac{\sqrt{I_1^{2} + I_2^{2}} \,.\, e^{i\theta}}{2\sqrt{\omega A}} \tag{7}$$

Here $\theta = \tan^{-1}(I_2/I_1)$ is the phase lag and I_1 and I_2 are Fresnel integrals given by

$$I_1 = \int_0^{2\sqrt{\omega A}} \cos(\pi/2\, u^{2})\, du$$

$$I_2 = \int_0^{2\sqrt{\omega A}} \sin(\pi/2\, u^{2})\, du$$

A is the dimensionless motion parameter equal to $Bt_e^{2}/\lambda F$. Here B is defined by the equation of parabolic motion, t_e is the total exposure time, λ is the wavelength of the incident light and F is the f number of the optical system.

3. RESULTS AND DISCUSSIONS

Figures 1 (*a*) to 1 (*c*) show the intensity distribution in the diffraction images of square wave targets for $N = 3$, 7 and 15 for various values of A. Similar curves have been plotted in case of sinewave objects and are illustrated in Figs. 2 (*a*) to 2 (*c*). Image in the absence of motion, *i.e.*, ($A = 0 \cdot 0$) are shown in all figures by dotted

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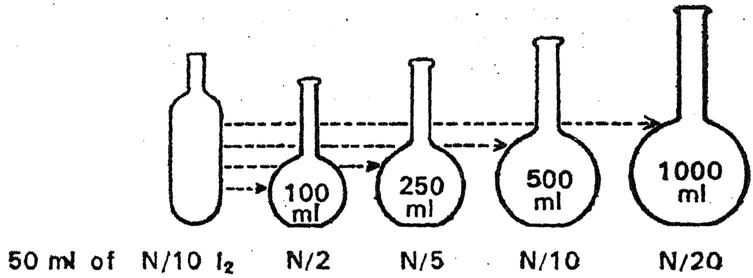

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DIFFRACTION IMAGES OF TRUNCATED PERIODIC OBJECTS IN PANORAMIC CAMERAS IN THE PRESENCE OF PARABOLIC IMAGE MOTION

A. K. GUPTA AND K. SINGH*

Department of Physics, Indian Institute of Technology, New Delhi 110 029

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where 4 L is the spatial period. Both v and L are in reduced units.

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$C(\omega)$ for the diffraction limited circular aperture is given by

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$$I_2 = \int_0^{2\sqrt{\omega A}} \sin(\pi/2u^2)\,du.$$

A is the dimensionless motion parameter equal to $Bt_e^2/\lambda F$. Here B is defined by the equation of parabolic motion[7], t_e is the total exposure time, λ is the wavelength of the incident light and F is the f-number of the optical system.

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* To whom all correspondence may please be addressed.

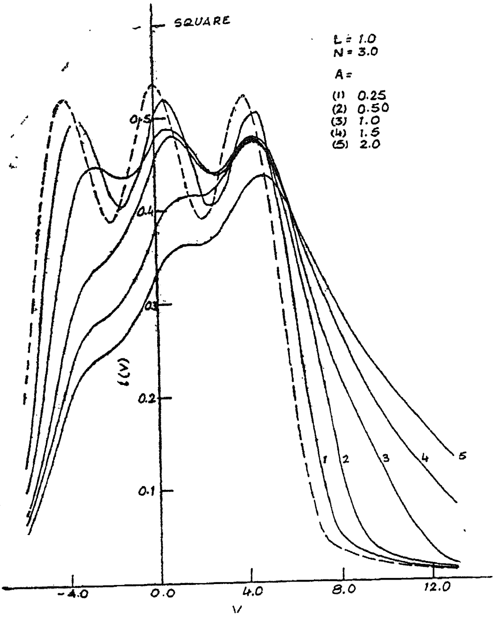

FIG. 1 (*a*). Diffraction Images of square wave object in presence of parabolic image motion. (A = 0·0, 0·25, 0·5, 1·0, 1·5 and 2·0, N = 3·0 and L = 1·0).

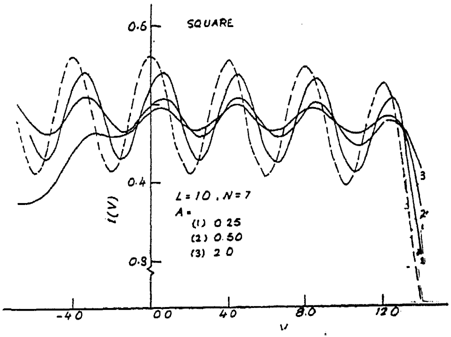

FIG. 1 (*b*). Same as 1 (*a*) with N = 7 and A = 0·0, 0·25, 0·50 and **2·0.**

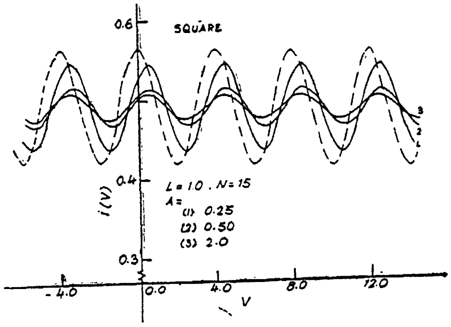

FIG. 1 (*c*). Same as 1 (*a*) with **N = 15 and** A = 0·0, 0·25, 0·50 and 2·0.

curves and these agree with the results published by Barakat and Lerman[2].

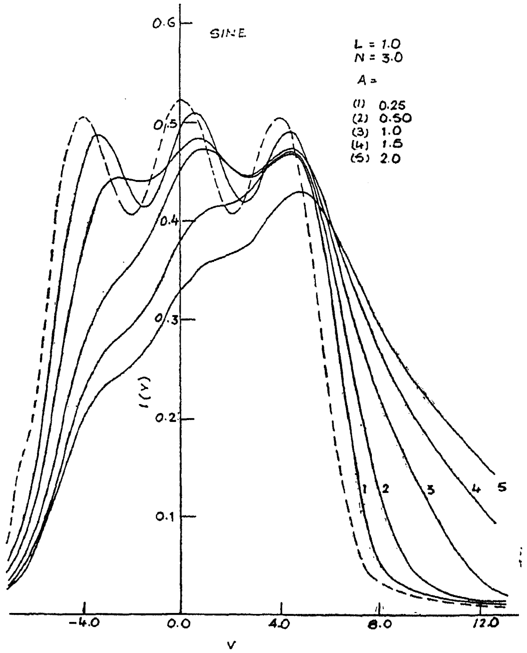

FIG. 2 (*a*). Diffraction images of sine wave object in the presence of parabolic image motion. (A = 0·0, 0·25, 0·5, 1·0, 1·5 and 2·0, N = 3·0 and L = 1·0).

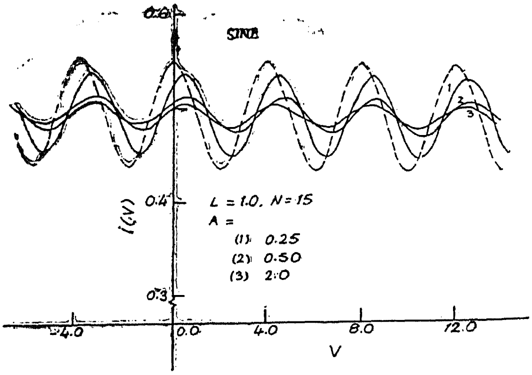

FIG. 2 (*b*). Same as 2 (*a*) with N = 7 and A = 0·0, 0·25, 0·5 and 2·0.

Square wave targets are imaged with higher contrast as compared to the sine wave targets. However, the general trend of results in the case of sine and square wave objects is very similar. It is evident from Figs. 1 and 2 that peak intensity position is shifted in presence of motion. The shift is, however, constant beyond A = 0·5. It is explained on the basis of the fact that the phase part of the transfer function becomes oscillatory beyond a certain value of ω for given value of

A (For example, at s = 0·2, for A = 1·0) and then becomes constant afterwards in high frequency region. It is also seen that the intensity distribution is asymmetric with respect to origin. Such asymmetry is also observed in investigating the comatic images by Inoue-tani, et al.[7], and Barakat[8].

It is obvious from Figs. 1 and 2 that the contrast is reduced in presence of parabolic motion, hence a degradation of image is caused. Degradation is increased as the value of A increases. The average irradiance in the central portion of the image is increased as N increases and approaches 0·50 as N approaches ∞. It must also be observed that there is no reversal of contrast for any value of A. There is no loss of periodicity for moderate values of A, e.g., for A = 0·5, 1·0 for N = 3 and 5. Some loss of periodicity results for higher values of A.

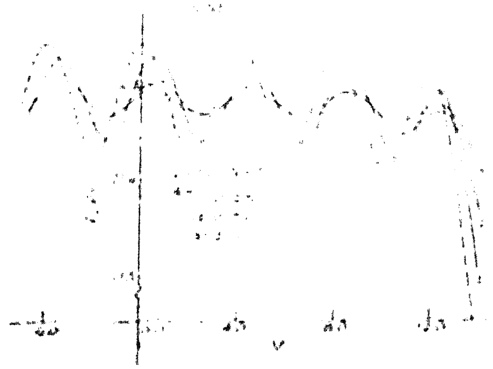

Fig. 2 (c) — Same as 2(a) with N = 15 and A = 0·0, 0·25, 0·5 and 2·0

Table I gives a better idea of the effect of motion on the image quality of targets under consideration. The method used for calculating modulation and average irradiance is as outlined in our previous papers[3,4]. It is inferred that the effect of image motion is practically nil on the average irradiance compared to the enormous changes in the modulation and hence modulation is ultimate tool to decide how many cycles are required for the approximation of an infinite sine wave target.

Additional Remarks

In this paper we confined ourselves to one dimensional objects. We would also like to mention that the motion degraded images of two dimensional objects such as disks, annulus, etc., cannot be calculated using Eqn. 6 for the OTF of the total system does not possess a rotational symmetry but acquires a certain azimuthal dependence. Work in this context has been done by Rattan and Singh[10].

TABLE 1

Modulation in the diffraction images of square and sine wave targets

A N	3	5	7	9	11	13	15
SQUARE WAVE							
0·0	1842	1518	1515	1504	1491	1495	1492
0·25	1136	1144	1142	1136	1128	1129	1123
0·50	0641	0479	0482	0484	0424	0446	0454
1·0		0·480	0484	0455	0446	0454	0451
1·5			0496	0496	0492	0496	0494
2·0			0299	0302	0299	0296	0301
SINE							
0·0	1189	1190	1188	1180	1170	1154	1172
0·25	0858	0895	0896	0894	0884	0882	0885
0·50	0339	0354	0377	0380	0372	0376	0377
1·0		0352	0355	0356	0349	0356	0355
1·5			0309	0312	0309	0312	0308
2·0			0233	0237	0238	0237	0236

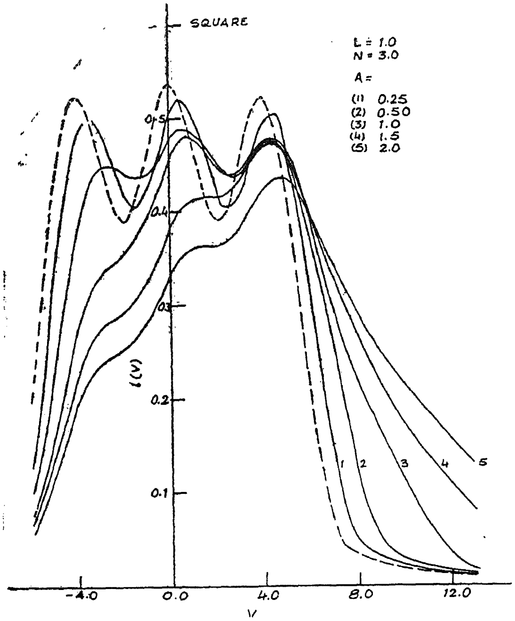

FIG. 1 (*a*). Diffraction Images of square wave object in presence of parabolic image motion. (A = 0·0, 0·25, 0·5, 1·0, 1·5 and 2·0, N = 3·0 and L = 1·0).

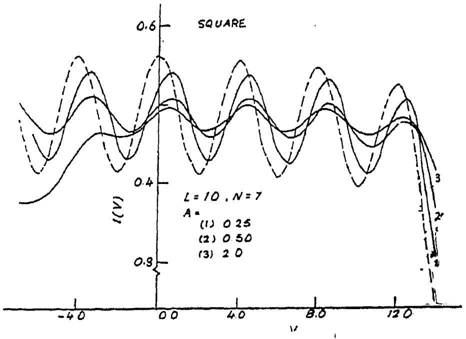

FIG. 1 (*b*). Same as 1 (*a*) with N = 7 and A = 0·0, 0·25, **0·50 and 2·0.**

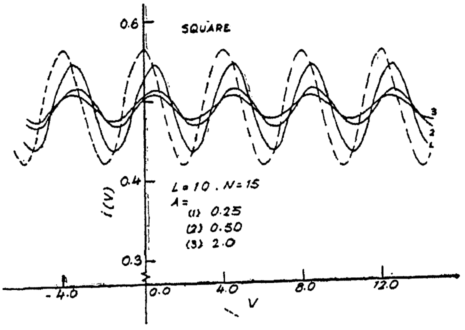

FIG. 1 (*c*). Same as 1 (*a*) with N = 15 and A = 0·0, 0·25, 0·50 and 2·0.

curves and these agree with the results published by Barakat and Lerman[2].

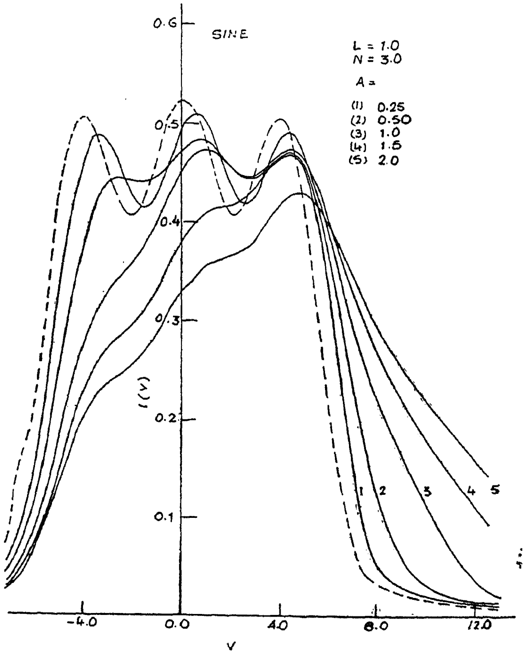

FIG. 2 (*a*). Diffraction images of sine wave object in the presence of parabolic image motion. (A = 0·0, 0·25, 0·5, 1·0, 1·5 and 2·0, N = 3·0 and L = 1·0).

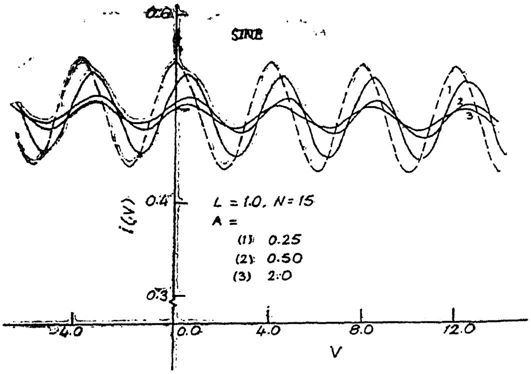

FIG. 2 (*b*). Same as 2 (*a*) with N = 7 and A = 0·0, 0·25, 0·5 and 2·0.

Square wave targets are imaged with higher contrast as compared to the sine wave targets. However, the general trend of results in the case of sine and square wave objects is very similar. It is evident from Figs. 1 and 2 that peak intensity position is shifted in presence of motion. The shift is, however, constant beyond A = 0·5. It is explained on the basis of the fact that the phase part of the transfer function becomes oscillatory beyond a certain value of ω for given value of

A (For examples at $\omega = 0\cdot7$, for $A = 1\cdot0$) and then becomes constant afterwards in high frequency region. It is also seen that the intensity distribution is asymmetric with respect to origin. Such asymmetry is also observed in investigating the comatic images by Ingelstam, *et al.*[17], and Barakat[18].

It is obvious from Figs. 1 and 2 that the contrast is reduced in presence of parabolic motion, hence a degradation of image is caused. Degradation is increased as the value of A increases. The average irradiance in the central portion of the image is increased as N increases and approaches 0·50 as N approaches ∞. It must also be observed that there is no reversal of contrast for any value of A. There is no loss of periodicity for moderate values of A, *e.g.*, for $A = 0\cdot5$, $1\cdot0$ for $N = 3$, and 5. Some loss of periodicity results for higher values of A.

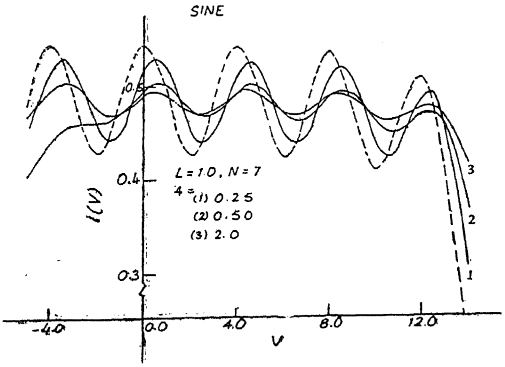

FIG. 2 (*c*). Same as 2 (*a*) with $N = 15$ and $A = 0\cdot0$, $0\cdot25$, $0\cdot5$ and $2\cdot0$.

Table I gives a better idea of the effect of motion on the image quality of targets under consideration. The method used for calculating modulation and average irradiance is as outlined in our previous papers[3,4]. It is inferred that the effect of image motion is practically nil on the average irradiance compared to the enormous changes in the modulation and hence modulation is ultimate tool to decide how many cycles are required for the approximation of an infinite sine wave target.

Additional Remarks :

In this paper we confined ourselves to one-dimensional objects. We would also like to mention that the motion degraded images of two-dimensional objects such as disks, annulas, etc., cannot be calculated using Eqn. 6 for the OTF of the total system does not posses a rotational symmetry but acquires a certain azimuthal dependence. Work in this context has been done by Rattan and Singh[19].

TABLE I

Modulation in the diffraction images of square and sine wave targets

SQUARE WAVE

A/N	3	5	7	9	11	13	15
0 ·0	·1532	·1518	·1515	·1504	·1491	·1495	·1492
0 ·25	·1136	·1143	·1142	·1136	·1125	·1129	·1127
0 ·50	·0641	·0479	·0482	·0483	·0474	·0446	·0454
1 ·0	. .	0 ·450	·0453	·0455	·0446	·0454	·0451
1 ·5	. .	. .	·0396	·0396	·0392	·0396	·0394
2 ·0	. .	. .	·0299	·0302	·0299	·0296	·0301

SINE

A/N	3	5	7	9	11	13	15
0 ·0	·1189	·1190	·1188	·1180	·1170	·1174	·1172
0 ·25	·0878	·0895	·0896	·0893	·0883	·0887	·0885
0 ·50	·0339	·0374	·0377	·0380	·0372	·0376	·0377
1 ·0	. .	·0352	·0355	·0356	·0349	·0356	·0355
1 ·5	. .	. .	·0309	·0312	·0309	·0312	·0308
2 ·0	. .	. .	·0233	·0237	·0235	·0237	·0236

It is also interesting to include the effect of lens-film combination on the diffraction images. It can be done by cascading the Film's MTF with the system MTF's provided the linear processes would be involved. However, complexity arises due to some non-linear phenomena involved during the exposure and development of the film.

We have considered in our calculation unit contrast truncated targets so that object contrast bears a linear relationship with image contrast. It is interesting to estimate the error caused by truncation by varying the modulation. For modulation less than unity there is non-linear relationship[20] between object and image contrast.

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1. Norton, C. L., *Photogramm. Eng.*, 1975, **41**, 203.
2. Barakat, R. and Lerman, S., *Appl. Opt.*, 1967, **6**, 545.
3. Singh, K. and Jain, N. K., *Nouv. Rev. Opt.*, 1972, **6**, 309.
4. —, Rattan, R. and Jain, N. K., *Appl. Opt.*, 1973, **12**, 1846.
5. —, Jain, N. K. and Rattan, R., *Atti. Fond. G. Ronchi.*, 1973, **28**, 545.
6. Nagel, M. R., *Evaluation of Motion Degraded Images*, Proc. of a Seminar held in Cambridge, Mass., Dec. 3–5, 1968, (NASA, Washington, DC. 1969).
7. Jensen, N., *Optical and Photographic Reconnaissance. System*, John Wiley and Sons, New York, 1968.
8. Dubovik, A. S., *Photographic Recording of High Speed Process*, Bergamon Press, Oxford, 1968.
9. Fouche', G., In : *Space Optics*, Eds., Maréchal, A. and Courtes, G., Gordon and Breach Science, Publ. N.Y., 1974, p. 374.
10. Rosenau, M. D., *J. Photogram. Eng.*, 1961, **27**, 421.
11. Som, S. C., *J. Opt. Soc. Am.*, 1971, **61**, 859.
12. Wolf, P. R., *Elements of Photogrammetry*, McGraw-Hill Book Co., N.Y., 1974.
13. Rattan, R. and Singh, K., *Atti. Fond. G. Ronchi.*, 1972, **27**, 727.
14. Gupta, P. C. and Singh, K., *Pramāṇa*, 1974, **3**, 390.
15. Jablonwski, D. P. and Singh, H., *Lee. Appl. Opt.*, 1973, **12**, 703.
16. Sawchuk, A. A., *Proc. IEEE*, 1972, **60**, 854.
17. Inglestam, E. *et al.*, *J. Opt. Soc. Am.*, 1956, **46**, 767.
18. Barakat, R. and Houston, A., *Ibid.*, 1965, **55**, 1142.
19. Rattan, R. and Singh, K., *Nouv. Rev. Opt.*, 1975, **6**, in press.
20. Jaiswal, A. K. and Bhusan, *J. Appl. Opt.*, 1974, **13**, 1839.

CHEMICAL COMPONENTS OF *ASPLENIUM LACINIATUM*

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ABSTRACT

Asplenium laciniatum has been found to contain octatriacontane, stearic acid, vitamin-K_3, phthiocol, β-sitosterol and β-sitosterol D-O-glucoside. The free sugars identified are sucrose (large amount), glucose, galactose and arabinose. This is the first report of the isolation of vitamin-K_3 and phthiocol from a plant source.

ASPLENIUM laciniatum is a fern belonging to the group of highly advanced cryptogens. Its alcoholic or acetone extract has a burning effect on the skin and produce a wound which takes about ten days to heal. Investigation of the chemical components was taken up in order to find out the active principles present in it.

The dried plant (4 Kg) obtained from Kalimpong (West Bengal) was exhaustively extracted with acetone and alcohol successively. Acetone from the acetone extract was removed under reduced pressure and the residue adsorbed on silica gel and chromatographed over a column of silica gel whereby the following compounds were obtained.

Compound A, eluted with petroleum ether, m.p. 68–70° (200 mg, from ethyl acetate). Its I.R. spectrum showed it to be aliphatic in nature. It did not answer Liebermann–Burchard test and moved along the solvent front on TLC in *n*-hexane. The mass spectrum showed a mass peak at m/e 534 and a cluster of peaks at 14 units ($-CH_2$) apart. The highest peak in each cluster represented C_nH_{2n+1} fragment which was accompanied by C_nH_{2n} and C_nH_{2n-1} peaks. Intense peaks were for C–4 and C–5 units and fragment intensity decreased in a smooth curve down to ($M^+-C_2H_5$). The (M^+-CH_3) peak was very weak. It was identified as octatriacontane.

Compound B, eluted with petroleum ether-benzene (1 : 3), m.p. 69–70° (60 mg, from methanol). Its methyl ester had m.p. 38–39°. Mass spectrum of the parent compound showed a parent peak at m/e 284. It was identified as stearic acid (octadecanoic acid).

Compound C, eluted with benzene, m.p. 104–105° (120 mg, yellow needles from petroleum ether). It gave positive test for a quinone. Its I.R. (1670 cm^{-1} for > C=O) and U.V. (λ_{max}^{EtOH} 246, 261 and 332 nm) spectra confirmed it to be a quinone. N.M.R. spectrum showed a doublet at δ 2·15 due to the presence of a quinonoid methyl group having an ortho H atom. A quartet at δ 6·8 integrating for one proton may be due to the ortho hydrogen. A multiplet at δ 7·8–8·15 integrated for four aromatic protons. In the mass spectrum it showed a parent peak at m/e 172 (100%). Appearance of abundant ions at m/e 144 (M^+–CO), 116 (M^+–2 CO) and at m/e 104 and its decomposition products at m/e 76 and 50 showed it to be a naphthoquinone with no benzenoid substituents[1,2]. From the above data the structure of the compound—C, could be given as 2-methyl 1 : 4-naphthoquinone which was confirmed by direct comparison with its synthetic sample (Co–TLC, m.m.p).

2-Methyl 1 : 4-naphthoquinone (menadione or Vit. –K_3) is known to occur among mammals[3]. Exhaustive survey of literature showed that it has not been isolated from nature. Vitamin K_3 has now been isolated for the first time from a plant source.

Compound D, eluted with benzene-chloroform (3 : 1), m.p. 173–74° (150 mg, yellow prisms from methanol). It gave positive test for a quinone. It also gave positive ferric chloride test indicating it to be phenolic in nature. Its I.R. (1660, 3410 cm^{-1} for > C=O and chelated –OH) and U.V. ($\lambda_{max}^{CHCl_3}$ 250, 280 and 335 nm) spectra confirmed it to be a hydroxy quinone. N.M.R. spectrum showed signals at δ 2·05 (3 H, $-CH_3$), δ 7·35 (1 H, $-OH$) and at δ 7·85–8·10 (m, 4 H, aromatic protons). Mass spectrum gave a parent peak at m/e 188 (100%). Presence of fragments at m/e 160 and 132 are due to the losses of one and two carbonyl groups respectively of the quinone. From the appearance of peaks at m/e 105 and 77 it could be concluded that the hydroxyl group in the naphthoquinone is at position two with no benzenoid substituents[1]. From the above data the structure of compound D, could be given as 2-hydroxy 3-methyl 1 : 4 naphthoquinone (phthiocol).

For further confirmation compound D was methylated by the dimethyl sulphate, potassium carbonate and acetone method and the methyl ether crystallised from methanol as light yellow needles, m.p. 93–94° (lit. 93°, from methanol). N.M.R. spectrum of the methyl ether showed an additional peak at δ 4·15 for the methoxyl group. I.R. spectrum lacks a peak at 3410 cm^{-1} (for the chelated hydroxyl). Its mass spectrum showed a parent peak as the base peak at m/e 202 along with normal fragmentation peaks for a naphthoquinone[1]. The structure of the compound D was confirmed by its synthesis[4] and its direct comparison with the synthetic sample (Co–TLC and m.m.p.).

Phthiocol is known to be a degradative product of the K-vitamins[5-8]. It is elaborated by bacteria[9-11] (micro-organism, unicellular) but its isolation has now been reported for the first time from a plant source (multicellular). The presence of vitamin-K_3 and pathiocol was also detected (TLC) in the fresh solvent extract of the fern and so they are not artefacts but exist as such in the plant.

Compound E, eluted with chloroform, had m.p. 136–38° (250 mg, from methanol), $[\alpha]_D$–40°. It gave positive Liebermann–Burchard and TNM tests, and ν_{max} 3,500 cm^{-1} (OH). It formed an acetate, m.p. 125–26° (from methanol). The compound was identified as β-sitosterol which was further confirmed by its direct comparison with an authentic sample (Co–TLC and m.m.p.).

Compound F, eluted with chloroform-methanol (95 : 5), m.p. above 300° and was almost insoluble in common organic solvents (100 mg). $[\alpha]_D$50° (pyridine). Acid hydrolysis gave β-sitosterol and D–glucose. Permethylation of Compound F, by Hakomori's method[12] and subsequent hydrolysis by Kiliani's reagent (HCl : AcOH ; H_2O ; 1 : 3·5 : 5·5) gave β-sitosterol and 2, 3, 4, 6-tetra-0-methyl D–glucose identified by paper chromatography. Enzymatic hydrolysis with emulsion showed β-linkage. The compound was identified as β-sitosterol-D-0-glucoside.

Alcohol extract on concentration deposited a white solid, m.p. 182–84° (30 g). It answered Molisch's test and gave an acetate, m.p. 68–69° (from methanol). The compound was identified as sucrose (Co–I.R.). Paper chromatography of the alcohol extract in butanol : pyridine : water ; 6 : 4 : 3 indicated the presence of glucose, galactose and arabinose (when sprayed with aniline hydrogen phthalate) as free sugars and confirmed by direct comparison with their respective authentic samples.

ACKNOWLEDGEMENT

The authors wish to thank Dr. A. K. Batta, Chemistry Department, A.R.S.D. College, Delhi University, for the synthetic sample of Vit. –K_3.

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1. Bowie, J. H., Cameron, D. W. and Williams, D. H., *J. Am. Chem. Soc.*, 1965, 87, 5094.
2. Di Mari, S. J., Supple, J. H. and Rapaport, H., *Ibid.*, 1966, 88, 1226.
3. Heilbron, I. and Hey, D. H. (Editorial Board), *Dictionary of Organic Compounds*, 1965, 4, 2248.
4. Fieser, L. F., *J. Biol. Chem.*, 1940, 133, 391.
5. Francis, J., Madinaveitia, J., Nacturk, H. M. and Snow, G. A., *Nature*, 1949, 163, 365.
6. Snow, G. A., *Congr. Intern, Biochim. Resumes Communs : 2 Cong. Paris*, 1952, page 95 (C. A. 1955, 49, 7648 *f*).
7. Burnett, A. R. and Thomson, R. H., *J. Chem. Soc.*, 1967, 21, 2100.
8. Thomson, R. H., *Naturally Occurring Quinones*, Academic Press, 2nd, Edition, 1971, p. 201.
9. Anderson, R. J. and Newman, M. S., *J. Biol. Chem.*, 1933, 101, 773.
10. Asano, M. and Takahashi. H., *J. Pharm. Soc. Japan*, 1945, 65 No. 3 A, 17.
11. Terni, M., *Boll. Soc. Ital. Biol. Sper.*, 1949, 25, 60 ; (C. A. 1951, 45, 2054 *f*).
12. Hakomori, S., *J. Biochem.*, 1964, 55, 205.

PALAEOMAGNETISM OF THE CENOZOIC VOLCANIC ROCKS FROM EAST AFRICA

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ABSTRACT

Palaeomagnetic results from 269 sites in the East African Cenozoic volcanics are presented. The post-Miocene volcanics were divided into three age groups, 0–1·8, 1·8–7·0 and 13–11 my. and their pole positions calculated. Along with the Miocene pole from Turkana, the Eocene-Oligocene pole from Ethiopia, the pre-Miocene post Karroo pole from Tororo and the mean Mesozoic pole from the rest of Africa, a polar wander path for Africa from Mesozoic to present is suggested.

The small pole displacements of the late Tertiary volcanics are both "far-sided and right-handed". Whereas this could be explained on the basis of Wilson's off-set dipole hypothesis, the much larger displacements shown by the older rocks must be attributed either to polar wander or to the drift of the African plate or to both.

DURING the last ten years the palaeomagnetic research group at the University of Nairobi has carried out detailed studies of a very wide spectrum of Cenozoic volcanics of East Africa. Results of a palaeomagnetic study of the Miocene lavas from Turkana, West of Rudolf form part of an unpublished thesis by Raja[1]. A detailed account of the palaeomagnetism of the volcanics of Central East Africa is given by Reilly[2], in his unpublished thesis. Patel and Gacii[3], have presented the results of their study on the Kapiti phonolites of Kenya. The palaeomagnetism of the Ethiopian flood basalts, believed to be Eocene–Oligocene in age has been reported by Brock *et al.*[4]. Recently Raja and Vise[5], have presented the results of their study on the Carbonatite volcanics of Tororo, S. E. Uganda, which are believed to be "pre-Miocene, post-Karroo" by Davies[6].

This paper presents a summary of the above results.

Rocks of age in the range 13–0 my., were divided into three groups A, B and C. The groups C/B boundary was chosen as 1·8 my. which corresponds to the Plio-Pleistocene boundary of the scale suggested by Berggren[7]. The relatively quiet period of volcanism 11–7 my. enabled the division between groups A and B. The Turkana lavas range in age from 32–13 my.[8]. But most of the basalts showed ages between 23 and 14 my. with a concentration at about 17 my.[9]. Hence the approximate age of 17 my. is assigned to the Turkana lavas considered as one unit.

Table I gives the mean directions and poles for all the groups mentioned above. Along with the mean Mesozoic pole for Africa at 261° E, 65° N[10], it is possible to suggest a polar wander path for Africa from Mesozoic to present. Such a path is shown in Fig. 1. It can be seen that the pole from the Tororo ring complex (TU) fills a wide gap between the Mesozoic pole and the East African Tertiary poles. The close similarity between the Australian polar wander curve for the Cenozoic and the African polar wander curve for the same period enabled McElhinny *et al.*[11], to suggest the existence of a common polar wandering component.

TABLE I

[illegible]

Group	Approximate age	Number of sites N	Direction D	I		α95*	Pole Longitude °E	Latitude °N	K	A95
C	1·8–0 m.y.	54	[illegible]	[illegible]	[illegible]	[illegible]	102·0	[illegible]	[illegible]	[illegible]
B	7–1·8 m.y.	102	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
A	13–11 m.y.	22	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	6·1
Turkana	Miocene ~ 17 m.y.	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
Ethiopia	Eocene oligocene	50	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
Tororo	Pre-Miocene	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]

D, Declination; I, Inclination; K, [illegible]
α95, A95, radius of 95% circle of confidence.
* Calculated for specimens.

TABLE II

[illegible]

Group	Approximate age	Number of sites N	Direction D	I		α95*	Pole Longitude °E	Latitude °N	K	A95
C_1 (Brunhes)	0·7 m.y.	24	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
C_2 (Matuyama)	0·7 m.y.	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	4·9

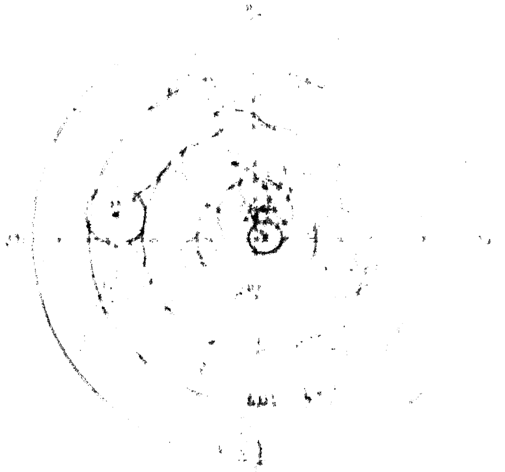

FIG. 1. Polar wander path for Africa from mesozoic to present. C, B, A, Poles from East Africa of age groups 1·8–0, 7–1·8 and 13–11 m.y. respectively; Mi, Miocene (~17 m.y.) Pole from Turkana; E, Eocene-oligocene pole from Ethiopia; T, Pre-miocene pole from Tororo (Uganda); M, mean mesozoic pole. (All poles are shown with 95% circle of confidence.)

However, Wilson has recently pointed out that the polar wander hypothesis need not be invoked to explain the [illegible] displacements of the upper Cenozoic poles. He has shown that the deviations are greatly reduced if the geocentric dipole is displaced northward along the spin axis (What is known as Wilson's offset dipole hypothesis). In a recent paper, Wilson and McElhinny have shown that if the northward offset of the dipole has existed, then the offset has been decreasing over late Tertiary time and is [illegible] approaching a small value during Quaternary. Our results as indicated in Fig. 1 and Table I are consistent with this finding.

ACKNOWLEDGEMENT

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1. Bowie, J. H., Cameron, D. W. and Williams, D. H., *J. Am. Chem. Soc.*, 1965, 87, 5094.
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TABLE I

Site mean directions, poles and statistical parameters

Group	Approximate age	Number of sites N	Directions				Poles			
			D°	I°	*k*	α_{95}°	Longitude °E	Latitude °N	K	A_{95}°
C	1·8–0 my.	54	1·3	−2·4	18·3	4·7	104·0	88·7	37·8	3·2
B	7–1·8 my.	102	3·3	−4·8	22·0	3·1	147·6	86·5	37·8	2·3
A	13–11 my.	22	1·7	−4·2	13·4	8·8	186·6	86·5	28·8	6·1
Turkana	Miocene ~17 my.	62	4·1	1·7	35·0	3·1	163·3	84·8	69·9	2·3
Ethiopia	Eocene-oligocene	20	7·2	7·2	34·9	5·6	167·7	80·8	59·3	4·3
Tororo	Pre-Miocene	3	4·5	−24·2	157*	9·9	195·5	75·8	172·2	9·4

D—Declination, I—Inclination, *k*, K—Fisher's precision parameter,
α_{95}, A_{95}—radius of 95% circle of confidence.
* Calculated for specimens.

TABLE II

Sub-division of group C into brunhes and matuyama age groups

Group	Approximate age	Number of sites N	Directions				Poles			
			D°	I°	*k*	α_{95}°	Longitude °E	Latitude °N	K	A_{95}°
C_1 (Brunhes)	<0·7 my.	24	2·5	−1·7	14·9	7·9	107·1	87·6	32·6	5·3
C_2 (Matuyama)	>0·7 my.	23	359·1	−1·0	19·7	7·0	353·8	88·4	38·5	4·9

However, Wilson[12] has recently pointed out that the polar wander hypothesis need not be invoked to explain the small displacements of the upper Cenozoic poles. He has shown that the deviations are greatly reduced if the geocentric dipole is displaced northward along the spin axis. (What is known as Wilson's offset dipole hypothesis). In a recent paper, Wilson and McElhinny[13], have shown that "if the northward offset of the dipole has existed, then this offset has been decreasing over late Tertiary time and is (within errors) approaching a small value during Quaternary". Our results as indicated by Fig. 1 and Table I are consistent with this finding.

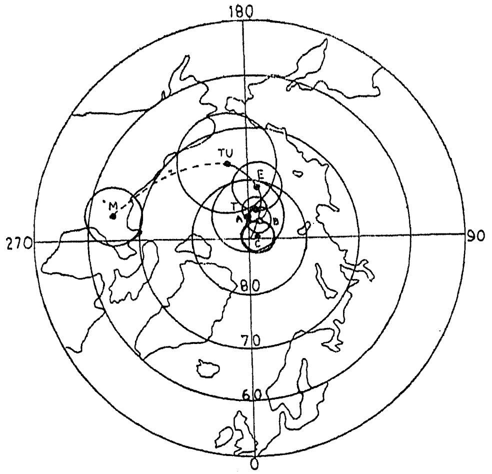

FIG. 1. Polar wander path for Africa from mesozoic to present. C, B, A, Poles from East African volcanics of age groups 1·8–0, 7–1·8 and 13–11 my. respectively; T, Miocene (~ 17 my.) Pole from Turkana; E, Eocene-oligocene pole from Ethiopia; TU, Pre-miocene pole from Tororo (Uganda); M, mean mesozoic pole; (All poles are shown with 95% circle of confidence).

Recently Watkins[14], analysed Brunhes epoch data from sites in the Indian Ocean and noticed that whereas the northern hemisphere data are consistent with the offset dipole hypothesis, the southern hemisphere data are consistent with the centred dipole hypothesis. Table II shows the subdivision of Group C into Brunhes and Matuyama. It can be seen that the poles for these groups are indistinguishable from the present. They are not only not "far sided", but if anything, "near sided" and as such, consistent with the centred dipole hypothesis which agrees with Watkins'[14], findings.

Table I shows that pole displacement of 5° in the Miocene increases to 9° in the Eocene and to 14° for the Tororo volcanics. If the small pole displacements of the late Tertiary and younger volcanics could be explained by the offset dipole hypothesis, the larger displacements shown by the older rocks must be attributed either to polar wander or to the drift of the African plate or to both.

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1. Raja, P. K. S., "A Palaeomagnetic study of some East African Rocks, *Ph.D. Thesis*, University of London, 1968.
2. Reilly, T. A. "Some Palaeomagnetic results from Central East Africa and the Seychelles Islands, *Ph.D. Thesis*, University of London, 1970.
3. Patel, J. P. and Gacii, P., *Earth Planet. Sci. Letters*, 1972, 16, 213.
4. Brock, A., Gibson, I. L. and Gacii, P., *Geophys. J. Roy Astron. Soc.*, 1970, 19, 485.
5. Raja, P. K. S. and Vise, J. B., *Earth Planet. Sci.* Letters, 1973, 19, 438.
6. Davies, K. A., *Memoir. No. VIII*, Geological Society of Uganda, 1956.
7. Berggren, W. A., *Nature*, 1969, 224, 1072.
8. Reilly, T. A., Mussett, A. E., Raja, P. K. S., Grasty, R. L. and Walsh, J., *Ibid.*, 1966, 210, 918.
9. Baker, B. H., Williams, L. A. J., Miller, J. A. and Fitch, F. J., *Tectonophysics*, 1971, 11, 191.
10. McElhinny, M. W., Briden, J. C., Jones, D. I. and Brock, A., *Rev. Geophy.*, 1968, 6, 201.
11. —, and Wellman, P., *Earth Planet. Sci. Letters*, 1969, 6, 198.
12. Wilson, R. L., *Geophys. J.R. Astr. Soc.*, 1970, 19, 417.
13. —, and McElhinny, M. W., *Ibid.*, 1974, 39, 570.
14. Watkins, N. D., *Ibid* 1972, 28, 193.

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LETTERS TO THE EDITOR

USE OF RADICAL SCAVENGERS IN X-RAY DATA COLLECTION FROM PROTEIN CRYSTALS

OCCURRENCE of radiation damage is a feature normally encountered while collecting X-ray intensity data from single crystals of proteins. It is well-known that protein crystals contain large solvent regions consisting mainly of water. The protein crystals, like the molecules themselves, are stable only in aqueous medium and are therefore mounted with the solvent in a capillary for data collection. According to the indirect action theory of radiation effects, the damage to the protein molecule is associated with the production of free radicals from the aqueous medium. These radicals, in turn, transfer the energy acquired from the incident radiation to the protein molecule. The protein molecule is thus left in an unstable, ionized state leading to significant changes in its structure and properties. The onset of radiation damage is detected by reduction in the intensity of the diffraction pattern which proceeds on to its total disappearance on prolonged irradiation. The amount of radiation dosage required to cause the damage varies with substances, some affected within hours of irradiation and some others within few days. Hence, most often, in protein crystallography, one is compelled to use several crystals to collect a complete set of data. Most proteins are available only in small quantities and it is often difficult to grow a large number of good crystals. Also, correlation of different data sets sometimes poses problems when several crystals are used for data collection.

Some of the procedures adopted for alleviating the effects of radiation damage to protein crystals are given below:

(*i*) *Fast data collection*: The speed of intensity measurement can be improved by the simultaneous measurement of several reflections, either photographically[1] or by the use of multiple counters[2] or position sensitive detectors[3,4].

(*ii*) *Data collection at low temperature*: Even though protein crystals may be expected to be more stable at low temperatures, ice formation in the solvent regions and the consequent enhancement in the volume often disrupts the protein crystal structure. However, by working at high pressure and low temperature, the more suitable ice III-ice IX region of the water-ice phase diagram which does not involve any volume change could be chosen[5]. Also, by the choice of a proper solvent which on freezing forms a glassy medium instead of ice, the volume increase could be avoided[6].

We here suggest the use of radioprotectants as a means for controling radiation damage to protein crystals. A large number of substances are known to be potential radioprotectants. The protecting machanism of some of these compounds, in particular, the aminothiol group of protectants, has been attributed to their ability to react with the radiation induced free radicals. Therefore, if the radioprotectants are introduced in the protein crystal, radiation damage could be effectively reduced since the protectants compete for the radiation-induced free radicals. The free radicals are prevented from reaching the protein molecules as they are scavenged by the radioprotectant immediately after they are produced. Also, the radioprotectant after reacting with the radiation-induced free radicals do not themselves react with the protein crystal[7]. The radioprotectant could be easily diffused into the solvent region of the protein crystals by dissolving them in the mother liquor, during crystallization. The introduction of the radical scavengers in the protein framework is not expected to produce significant changes in the diffraction pattern of the crystal, as the former are generally low molecular weight compounds.

Materials Science Division,
National Aeronautical
Laboratory,
Bangalore 560 017,
November 21, 1975.

S. RAMASESHAN.
KALYANI VIJAYAN.

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1. Arndt, U. W., Champness, J., Phizackerley, P. and Wonnacott, A. J., *J. Appl. Cryst.*, 1973, 6, 457.
2. Phillips, D. C., *J. Sci. Inst.*, 1964, 41, 123.
3. Prince, E. and Santoro, A., *Acta Cryst.*, 1975, 31 A, Part 3 Supplement, pp. S 234.
4. Hamlin, R., Cork, C., Nielson, C., Vernon, W. and Xuong, Ng. H., *Ibid.*, p. S 234.
5. Thomanek, U. F., Parak, F., Mossbauer, R. L., Formanek, H., Schwager, P. and Hoppe, W., *Acta Cryst.*, 1973, 29 A, 263.
6. Haas, D. J. and Rossmann, M. G., *Acta Cryst.*, 1970, 26 B, 998.
7. Casarett, A. P., *Radiation Biology*, Prentice Hall, Inc., Englewood Cliffs, New Jersey, 1968, p. 258.

IRON, SODIUM AND POTASSIUM CONTENTS TO AID ESTIMATION OF THE RELATIVE CONTRIBUTIONS OF VARIOUS SOURCES TO THE ATMOSPHERIC AEROSOLS—A PROPOSITION

INPUT of tropospheric aerosols—which amount to about 10^7 tons day^{-1}, can broadly be classified as natural and man-made. In an exhaustive review Hidy and Brock[1] have discussed various sources and their global contributions to the tropospheric aerosols. An initial attempt was made at this laboratory[2] to estimate the relative contributions of natural and man-made sources to atmospheric aerosol formation by taking account of their Fe and K contents only, which is perhaps not valid where sea spray also plays a vital part. In the present note Na is also taken into account in addition to Fe and K to take care of the oceanic contribution.

If one restricts interest to industrial urban areas only, it may perhaps be justified to assume that only the following shall be the principal sources of atmospheric aerosols: (1) sea spray–more so in coastal regions, (2) dust rise by winds, construction and building operations, (3) combustion of fuels–mainly coal–and other industrial operations and (4) oxidation of H_2S and SO_2 to sulphates and probably other sulphur containing organic compounds which produce aerosols free from Fe, Na, and K. It may as well be noted that aerosols arising from sources (1), (2), and (3) will contain Fe, Na and K in widely different proportions. In non industiral areas the contribution from (3) will be relatively much less.

We now make the following reasonable assumptions: (*a*) That the major portion by weight of the terrestrial dust is of local origin–say within a few tens of Km., and (*b*) That the source of coal supply to given region remains the same over reasonable periods of time and thus their relative contents of Fe, Na and K remain more or less constant.

From literature we find that for the terrestrial dust and sea spray the global Fe/K ratios are 1·92 and 0·00 respectively and the Na/K ratios for the same are 1·1 and 30 respectively. For bituminous coal and lignite also as some measurements indicate[3] the above two ratios are very much different. It is, therefore, surmised that the widely different ratios Fe/K and Na/K in the three major contributors to the tropospheric aerosols will give rise to unique ratios of Fe/K and Na/K in a composite sample of atmospheric dust depending upon the relative contributions from these sources.

We now mathematically show how the estimation of Fe/K and Na/K in suspended dust samples can lead to the evaluation of the contributions from the above three major sources.

We know that the Fe/K and Na/K ratios in sea spray are 0·0 and 30·0 respectively. Let the measured ratios of Fe/K in the local soil and fly-ash samples be A and B respectively and the ratios of Na/K in the same be C and D respectively. If the fractions of K contributed by the soil, the fly-ash and the sea-spray are *a*, *b* and *c*, respectively then.

$$a + b + c = 1 \qquad (1)$$

The observed ratio Fe/K for the suspended dust load will be given by

$$\mathrm{X} = \frac{a\cdot \mathrm{A} + b\cdot \mathrm{B} + \mathrm{C}\cdot 0}{a + b + c} \quad (\text{Fe/K for sea-spray} = 0)$$

from (1) this reduces to

$$\mathrm{X} = a\cdot \mathrm{A} + b\cdot \mathrm{B} \qquad (2)$$

In the same way the ratio Na/K in the suspended dust sample is given by

$$\mathrm{Y} = a\cdot \mathrm{C} + b\cdot \mathrm{D} + c\cdot 30 \qquad (3)$$

(Na/K for sea-spray = 30)

combining equations (1), (2) and (3) we get

$$a = \frac{\mathrm{B}\,(30 - \mathrm{Y}) - \mathrm{X}\,(30 - \mathrm{D})}{\mathrm{B}\,(30 - \mathrm{C}) - \mathrm{A}\,(30 - \mathrm{D})} \qquad (4)$$

$$b = (\mathrm{X} - a\cdot \mathrm{A})/\mathrm{B} \qquad (5)$$

and $c = 1 - (a + b)$ (6)

Since for a given region A, B, C and D are assumed to be constants and predetermined, a direct measurement of x and y will enable us to evaluate a, b and c and thus total material contributed from the three major sources mentioned earlier with the help of their known contents, of K. The sum of the materials contributed by these sources when subtracted from the total mass of the suspended dust sample gives the contribution from the fourth category of sources mentioned earlier which are free from Fe, Na and K.

It is expected that in most cases, the present method will give a fairly good estimate of the man-made and natural contributions to the atmospheric dust load, although in certain cases account shall have to be taken for specific sources such as the cement dust, etc.

Division of Radiological Protection, P. K. ZUTSHI.
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1. Hidy, G. M. and Brock, J. R., *An Assessment of the Global Sources of Tropospheric Aerosols*, 2nd Clean Air conference, International Union of Air Pollution Prevention Association, Washington, D.C., (Dec. 1970).
2. Zutshi, P. K. Sequeria, R. and Banerji. T., *J. Instt. Engs. Durgapur*, (India), 1969, 6, 42.
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$$= \frac{1}{C_M}[aC_A + H^+ - A - A^-] \qquad (1)$$

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In the present investigation the stability constants of VO (II) and UO_2 (II) with 2-nitro 1-naphthol and 2-nitroso 1-naphthol chelates were compared with log K values of the other bivalent metal ions. According to the sequence the order stability between two oxo-cations is VO (II) > UO_2 (II). Such an order of the stability has been earlier observed in VO (II) and UO_2 (II)—poly hydroxy aromatic chelates[11]. However the positions of VO (II) and UO_2 (II) in comparison with other bivalent metal ions have been different in different investigations. Mahadevan *et al.*[6], have found that the stability con-

TABLE I

The stability constants of the metal chelates of 2-nitro 1-naphthol and 2-nitroso 1-naphthol with some bivalent metal ions at 35 ± 0·1° C

Metal ion	2-nitro 1-napthol log K_1	2-nitroso log K_1	1-naphthol log K_2
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Mn (II)	3·08	3·91	3·53
Co (II)	3·24	7·63	4·34
Ni (II)	3·40	6·65	..
Cu (II)	3·79	7·50	5·42
Zn (II)	2·82	5·95	..
VO (II)	5·29	7·24	..
UO_2 (II)	5·21	7·09	..

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Where the symbols have the usual significance. The $\bar{n}$ values obtained at different pH values are plotted against pA^- for the metal ions. The stability constants K_1 and K_2 which are equivalent to pA^- at $\bar{n} = 0 \cdot 5$ and $\bar{n} = 1 \cdot 5$ are taken from the formation curve.

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The stability constants of the metal chelates of 2-nitro 1-naphthol and 2-nitroso 1-naphthol are given in Table I. The order of stability constants of bivalent transition metal ions can be explained in terms of crystal field stabilization energy. There is a gradual rise in the crystal field stabilization energy of the series Mn (II) to Ni (II) aquo complexes or complexes with the weak field ligands. The stability order from Mn (II) to Ni (II) can therefore be attributed to the gradually increasing C F S E values. Though the crystal field stabilization energy for Cu (II) is lower than Ni (II), the complex stability order is Cu (II) > Ni (II). This anamoly has been explained as a sequence of Jahn–Teller stabilization energy in the case of Cu (II) ion. This may lead to the conclusion that the ligands in the present investigation are perhaps weak field ones. The higher stability of Co (II)—2-nitroso 1-naphthol chelate as compared with that of Ni (II)—2-nitroso 1-naphthol chelate may be attributed to the higher oxidation state of cobalt, Co (III) in the chelate. The oxidation of Co (II) to Co (III) might have taken place during the complex formation.

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VO (II)	5·29	7·24	..
UO_2 (II)	5·21	7·09	..

In the present investigation the stability constants of VO (II) and UO_2 (II) with 2-nitro 1-naphthol and 2-nitroso 1-naphthol chelates were compared with log K values of the other bivalent metal ions. According to the sequence the order stability between two oxo-cations is VO (II) > UO_2 (II). Such an order of the stability has been earlier observed in VO (II) and UO_2 (II)—poly hydroxy aromatic chelates[11]. However the positions of VO (II) and UO_2 (II) in comparison with other bivalent metal ions have been different in different investigations. Mahadevan *et al.*[6], have found that the stability constant of Cu (II)—1-nitroso 2-naphthol is greater than that of UO_2—1-nitroso 2-naphthol; on the contrary Jefurkar *et al.*[11], observed a stability order UO_2 (II) > Cu (II). In the present investigations the stability order of Cu (II) > UO_2 (II) in case of 2-nitroso 1-naphthol chelates and Cu (II) < UO_2 (II) in the case of 2-nitro 1-naphthol chelates. In this context Back[12] has rightly pointed out that the natural stability order of Irving–Williams is probably valid only for the high spin complexes and the stability order might even become less defined with oxo cations. The low stability of the metal chelates of 2-nitro 1-naphthol as compared with 2-nitrose 1-naphthol chelates is due to the fact that the oxygen of a resonating nitro group is weaker basic centre than the oxygen of the nitroso group.

One of the authors (P.I.), is grateful to U.G.C. for providing financial assistance.

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---

1. Jonassen, H. B. and Monsabert, W. R., *J. Am. Chem. Soc.*, 1952, **74**, 5298.
2. Van Uitert, L. G. and Fernelius, W. C., *Ibid.*, 1954, **76**, 375.
3. Dyrssen, D., Dyrssen, M. and Jonassen, E., *Acta Chem. Scand.*, 1956, **10**, 106.
4. Callahan, C. M., Fernelius, W. C. and Block, B. P., *Anal. Chim. Acta*, 1957, **16**, 101.
5. Rydberg, J., *Acta. Chem. Scand.*, 1960, **14**, 157.
6. Mahadevan, M., Shetty, S. Y. and Sathe, R. M., *Indian J. Chem.*, 1971, **9**, 258.
7. Hoffman, O., *Ber.*, 1885, **18**, 46.
8. Schwarzenbach, G., *Complexometric Titrations*, Interscience, New York, 1957, p. 77.
9. Van Uitert, L. G. and Haas, C. G., *J. Am. Chem. Soc.*, 1953, **75**, 451.
10. Calvin, M. and Wilson, K. W., *Ibid.*, 1945, **67**, 2003.
11. Jefurkar, C. R., Mavani, I. P. and Bhattacharya, P. K., *Indian J. Chem.*, 1972, **10**, 1190.
12. Beck, M. T., *Chemistry of Complex Equilibria*, Van Nostrand–Reinhold Co., New York, 1970, p. 251.

VINYL POLYMERIZATION BY THALLIUM (III) SALTS

THE mechanism of oxidation of organic substrates by thallium salts has been fairly well established[1,2]. Little is known about the reaction of thallium with radicals[3]. But for a solitary report there has not been much evidence for the reaction between Tl (III) and any free radical[4]. The use of thallium (III) salts as polymerization catalysts has been scant. A few patents have been obtained on the gas phase olefin polymerization[5]. However the use of thallium (III) salts as catalysts for vinyl polymerization in solution has not been reported till now. For the first time the present study reports several interesting aspects of thallium (III) acetate catalyzed polymerization of acrylamide.

Thallium (III) acetate is a fairly efficient initiator causing a 50% conversion in 30 minutes at a temperature of 50° C. The rate of acrylamide polymerization was found to be directly proportional to the monomer concentration (*vide* Fig. 1). Another interesting feature which was observed was that the rate of polymerization remained constant with change in the metallic salt concentration, even when the thallium (III) acetate concentration was changed hundredfold, *viz.*, from 5×10^{-5} to 5×10^{-3} M. The rate of decomposition of the thallium (III) acetate however varied linearly with the concentration of the metallic salt indicating the possibility of simultaneous reaction paths available for the thallium (III) salt (*vide* Fig. 1). Chain termination by the thallium (III) acetate is postulated to explain the kinetics.

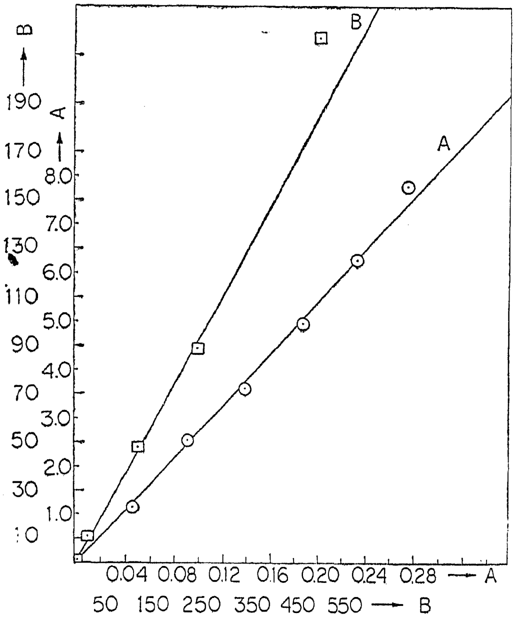

FIG. 1. Effect of Monomer concentration and metal concentration on Rates. T = 50° C; t = 30 min. A, $-d[M]/dt \times 10^5$ vs. [M]; B, $-d[I]/dt \times 10^8$ vs. $[I] \times 10^5$.

$$Tl(OAc)_3 \xrightarrow{k_d} Tl(OAc) + 2\,AcO^\circ$$

$$AcO^\circ + M \xrightarrow{k_i} M^\circ$$

$$M^\circ + M \xrightarrow{k_p} M_2^\circ$$

. . . .

$$M_{n-1}^\circ + M \longrightarrow M_n^\circ$$

$$M_n^\circ + Tl(OAc)_3 \xrightarrow{k_t} \text{polymer}$$

Assuming the steady state conditions and assuming k_p and k_t to remain constant and independent of chainlength the following expressions are derived :

$$\frac{-d[\mathrm{M}]}{dt} = \frac{k_d k_p}{k_t}[\mathrm{M}]$$

and

$$\frac{-d[\mathrm{I}]}{dt} = 2k_d[\mathrm{I}]$$

A similar feature was observed in the photopolymerization of methacrylamide by trans-diazidotetramminecobalt (III) azide[6] and in the photopolymerization of N-vinylpyrrolidone by azidopentamminecobalt (III) chloride[7], wherein termination by the excited complex molecule was postulated. The rate constants calculated were $k_d = 2{\cdot}18 \times 10^{-4}$ sec^{-1} and $k_p/k_t = 1{\cdot}25$. The unusually low k_p/k_t value may be explained by the high termination constant for thallium (III) acetate.

Another interesting aspect found was that thallium (III) acetate is an excellent photoinitiator of vinyl polymerization ; as a matter of fact the efficiency being much more photochemically than thermally. Further work is in progress to unravel the ramifications of the kinetics.

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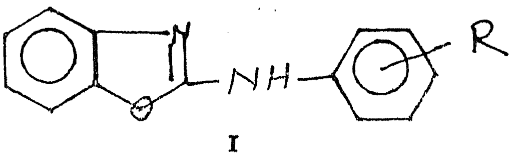
1. Balakrishnan, T. and Santappa, M., *Curr. Sci.*, 1973, 42, 418.
2. McKillop, A., Hunt, J. D., Naylor, R. D. and Taylor, E. C., *J. Am. Chem. Soc.*, 1971, 93, 4918.
3. Lee, A. G., *The Chemistry of Thallium*, Elsevier Publishing Co., London, 1971, p. 315.
4. Collinson, E., Dainton, F. S., Mile, B., Tazuke; S. and Smith, D. R., *Nature*, 1963, 198, 26.
5. Belg. 566, 530 ; Oct. 1958 ; Brit. 812, 176 April 22, 1959 ; Brit. 893, 304; July 1959 ; U.S. 3076794, Feb. 5, 1963.
6. Kothandaraman, H. and Santappa, M., *Polymer J.*, 1971, 2, 148.
7. —, Srinivasan, K. S. V. and Santappa, M., *J. Polymer. Sci.*, A–1, 1972, 10, 3685.

SYNTHESIS OF SUBSTITUTED 2-ANILINO-BENZOXAZOLES

Benzoxazoles have been reported to demonstrate diverse biological properties[1-5]. Some 3-substituted benzoxazolinones synthesised and screened by us have also shown antibacterial activity[3-5]. Syntheses of certain substituted 2-anilinobenzoxazoles (I) for biological screening have been given in this note.

For the synthesis of (I), 2-chlorobenzoxazole and aromatic amines in equimolar proportions were allowed to react in anhydrous benzene. 2-(4′-carbomethoxyanilino) benzoxazole (I *a*) obtained from methyl[4] aminobenzoate and 2-chlorobenzoxazole has been hydrolysed to the corresponding carboxylic acid (I *e*). I *e* was also obtained when 2-chlorobenzoxazole and 4-aminobenzoic acid were heated together. The product (I *j*) synthesised from 4-aminoacetophenone has been smoothly reduced to the corresponding secondary alcohol (I *k*) with sodium borohydride. The infrared spectrum of I *j* showed characteristic absorption peaks at 3270 cm^{-1} (NH) and 1678 cm^{-1} (C=O). Reduction of I *j* to I *k* caused the disappearance of carbonyl absorption from the i.r., of I *k* ; an additional band was observed at 3077 cm^{-1} (OH, broad) besides band at 3279 cm^{-1} (NH).



I

Biological Data.—All the compounds described in Table I were screened for their inhibitory effects against four microorganisms : *Staphylococcus aureus, Escherichia coli, Salmonella typhi,* and *Bacillus megaterium* by agar diffusion technique[6]. None of the compounds showed any significant activity.

Experimental

Melting points were determined in open capillaries and are not corrected. Infrared spectra were recorded on a Perkin–Elmer 337 Spectrophotometer in KBr.

Substituted 2-Anilinobenzoxazoles (I).—2-Chlorobenzoxazole (0·01 mole) and a suitable aromatic amine (0·01 mole) were suspended in 50 ml of dry benzene and refluxed for three hours. The contents assumed a light pink colour which slowly faded away as the reaction progressed. Excess of benzene was distilled off and the solid product thus obtained was recrystallised from ethanol. The analytical and other pertinent data are recorded in Table I.

Reduction of 2-(4′-Acetylanilino) Benzoxazole (Ij) to (I k) with Sodiumborohydride.—To a solution of I *j* (0·50 g) in ethanol was added gradually 0·2 g. of $NaBH_4$ dissolved in ethanol and the mixture heated on a waterbath for 1 hr. and the excess of ethanol distilled under reduced pressure ;

TABLE I

Substituted 2-Anilinobenzoxazoles (I)

Sl. No.	R	M.P. °C	Molecular formula	% Analyses Found	 Calcd.
I *a*.	4–COOCH$_3$	213–214	$C_{15}H_{12}N_2O_3$	N[d], 10·56	10·45
I *b*.	4–COOC$_2$H$_5$	203[b]	$C_{16}H_{14}N_2O_3$	..	..
I *c*[a]	4–COOC$_3$H$_7$	181–184	$C_{17}H_{16}N_2O_3$	N[d], 9·34	9·45
I *d*.	4–COOC$_4$H$_9$	150–151	$C_{18}H_{18}N_2O_3$	N, 8·94	9·02
I *e*.	4–COOH	>250[c]	$C_{14}H_{10}N_2O_3$	..	..
I *f*.	3–COOCH$_3$	166–167	$C_{15}H_{12}N_2O_3$	N, 9·94	10·45
I *g*.[a]	3–COOC$_2$H$_5$	197	$C_{16}H_{14}N_2O_3$	N[d], 9·72	9·92
I *h*.	2–COOCH$_3$	123–124	$C_{15}H_{12}N_2O_3$	N[d], 10·42	10·45
I *i*.	4–CH$_3$	176–178	$C_{14}H_{12}N_2O$	N[d], 11·98	12·49
I *j*.[a]	4–COCH$_3$	205	$C_{15}H_{12}N_2O_2$	N[d], 10·73	11·10
I *k*.[a]	4–CH (OH) CH$_3$	174–175	$C_{15}H_{14}N_2O_2$	N[d], 10·70	11·02
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a Infrared spectra were in accord with the assigned structures.
b Lit.[7] m.p. 202°.
c Lit.[7] m.p. 326–328°.
d Analysed correctly for C and H also.
Compounds (I *a*–I *l*) were obtained in 45 to 65% yields.

50 ml of water was then added to it. The contents were then extracted with ethyl acetate and the extract dried over $MgSO_4$. The residue obtained after removing ethyl acetate was recrystallised from ethyl acetate-petroleum ether (b.p. 60–80°).

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---

1. Martin, G. J. and Moss, J. N., *Am. J. Pharm.*, 1949, 121, 169.
2. Lespagnol, A., Mercier, J. and Lespagnol, C., *Arch. Int. Pharmacodyn.*, 1953, 94, 212; *Chem. Abstr.*, 1954, 48, 10003.
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5. —, and Nobles, W. L., *J. Pharm. Sci.*, 1968, 57, 39.
6. —, and —, *Ibid.*, 1968, 57, 1801.
7. Advani, S. B., *Ph.D. Dissertation, University of Mississippi*, 1967.

QUANTITATIVE SEPARATION OF TITANIUM FROM COPPER BY THIN LAYER CHROMATOGRAPHY

BINARY mixtures of titanium with copper, aluminium, iron, vanadyl, uranyl, nickel, cobalt, chromium and tin salts have been separated on plates coated with silica gel without binder. The solvent used was methanol-50% nitric acid; 9 : 1. The quantitative aspects of this separation were studied using 200 γ of copper and 198 γ of titanium. The separation was found to be quantitative.

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TABLE I
R_f values of 46 common metal ions in methanol-50% HNO_3; 9 : 1 system

Cations	R_f	Cations	R_f	Cations	R_f	Cations	R_f
Ag+	0·19	Cr+3	0·59	Mo+6	0·65	Sr+2	0·85
Al+3	0·68	Cs+	0·66	Mg+2	0·85	Te+4	0·55
As+3	0·35	Fe+2	0·64	Nb+5	0·02	Th+4	0·70
Au+3	0·43	Fe+3	0·62	Ni+2	0·60	Ti+4	0·06
Ba+2	0·88	Ga+3	0·82	Pb+2	0·66	UO_2+2	0·53
Be+2	0·68	Hg_2+2	0·22	Rb+	0·81	VO+2	0·63
Bi+3	0·72	Hg+2	0·57	Ru+3	0·60	Cu+2	0·76
Cd+2	0·81	Ir+4	0·74	Sb+2	0·36	W+6	0·02
Ca+2	0·66	K+	0·83	Se+4	0·57	Y+3	0·61
Ce+3	0·71	La+3	0·68	Sn+2	0·86	Zr+4	0·04
Co+2	0·80	Mn+2	0·68	Sn+4	0·69	Zn+2	0·77
Ce+4	0·89					Pt+4	0·55

Solutions (containing 190–200 γ) of titanium and other cations were spotted on the TLC plate in the form of a uniform streak. The plates were then dried and kept in the developing solvent methanol-50% nitric acid (9 : 1) till the solvent ascended about 12 cm. on the plate. Pilot chromatograms were run under similar conditions to ascertain the positions of the respective spots after development. Only titanium spot was found to be at the point of application. The R_f values for the 46 metal ions were determined (Table I). Binary mixtures of titanium with copper, aluminium and chromium were also subjected to separation by the above method. The portion containing titanium on the TLC plate was cut and removed by a spatula into a beaker. Titanium was recovered by leaching six times with 5% HCl solution. The combined washings were then reduced to 2 ml by evaporation on a waterbath. Titanium was then determined spectrophotometrically using sulphosalicylic acid[2]. The results obtained indicated that titanium can quantitatively be separated from copper while the separation of titanium in the other binary mixtures yielded low results.

The author expresses his gratitude to Dr. Mohsin Qureshi, Head, Department of Chemistry, Z. H. Engineering College, A.M.U., Aligarh, under whose guidance this work was carried out. He is also thankful to Smt. Gayatri Devi Varshney, for assistance.

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---

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Au+3	0·43	Fe+3	0·62	Ni+2	0·60	Ti+4	0·06
Ba+2	0·88	Ga+3	0·82	Pb+2	0·66	UO_2+2	0·53
Be+2	0·68	Hg_2+2	0·22	Rb+	0·81	VO+2	0·63
Bi+3	0·72	Hg+2	0·57	Ru+3	0·60	Cu+2	0·76
Cd+2	0·81	Ir+4	0·74	Sb+2	0·36	W+6	0·02
Ca+2	0·66	K+	0·83	Se+4	0·57	Y+3	0·61
Ce+3	0·71	La+3	0·68	Sn+2	0·86	Zr+4	0·04
Co+2	0·80	Mn+2	0·68	Sn+4	0·69	Zn+2	0·77
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The eye lenses of Rat (*Ratus ratus*), Order Rodentia ; Rabbit (*Oryctologus cemulus*), Order Lagomorpha ; Goat (Capra) and Buffalo (*Bubalus bubalus*), Order Artiodactyla ; Dog (*Canis familiaris*) and Cat (Domestic), Order Carnivora ; Monkey (*Macaca mulatta*), Order Primate were

TABLE I

The mineral contents in mammalian eye lenses

	Ca	Mg	Fe	Zn	Cu	Mn
	mg/100 g		← µg/100 g →			
Rat	3·5	0·48	580	—	—	—
Dog	5·6	4·1	68	1780·0	160·0	—
Cat	6·3	3·7	42	1535·0	148·0	—
Rabbit	3·8	0·51	670	18·3	4·1	8·1
Buffalo	4·9	0·96	106	216·0	216·0	27·3
Goat	4·3	1·02	119	246·0	290·0	20·1
Monkey	4·5	0·32	406	270·0	65·0	—

— not detected.

The values are calculated on the basis of wet weight of eye lenses and is the mean of four determinations each from a group consisting of 30–50 eye lenses.

selected for the present study as they belong to different orders of class mammalia.

The whole eye balls were obtained within two hours of the death of the animal. To minimize variation due to age, the eye balls from the adult animals were taken. They were carefully dissected and the lenses were freed from the capsule and the surrounding tissues on ice, blotted lightly to remove adhering moisture, dried in a vacuum desiccator to constant weight and ashed at 550° C. The acidified solution of ash was used for the estimation of calcium[1], magnesium[2], iron[3] copper[4], zinc[5] and manganese[6].

Results and Discussion

The results (Table I) do not show any consistent change in the order arranged, though variations are found among these species. The degree of such variations seems to be mainly dependent on the feeding habits of the animals. Magnesium, calcium and zinc are significantly high in carnivores as compared with the other animals, whereas the opposite correlation exists for iron content. Burdon-Cooper has reported traces of zinc in some species[7] and the absence of iron and copper in ash of individual lenses[8] but Carmi[9] has shown the presence of iron in traces. In the present study, iron was found in the eye lenses of all the species studied, being maximum in rabbit and rat eye lenses. Copper was totally absent in the rat but traces were found in the rabbit. Schlopak *et al.*[10] also confirmed the presence of traces of iron and copper in some of the animal lenses. Manganese was absent in dog, cat, rat and monkey but present in traces in rabbit, goat and buffalo eye lenses.

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1. Clark, E. P. and Collip, J. B., *J. Biol. Chem.*, 1925, 63, 461.
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3. Andrews, J. S. and Fett, C., *Cer. Chem.*, 1941, 18, 819.
4. Ventura, S. and King, E. J., *Biochem. J.*, 1951, 48, lxi.
5. Caughy, R. A., Holland, E. B. and Ritchie, W. S., *J. Assoc. Official Agric. Chemists.*, 1938, 21, 204.
6. Weise, A. C. and Johnson, B. C., *J. Biol. Chem.*, 1939, 127, 203.
7. Burdon-Cooper, J., *Concilium Ophthalmologicum*, 1929, 13, 185.
8. —, *Trans. Ophthal. Soc. U.K.*, 1928, 48, 340.
9. Carmi, A., *Boll d', Occul.*, 1929, 8, 1356.
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IMPROVEMENT OF THE NUTRITIVE VALUE OF TOMATOES

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Thus, the β-carotene content of the normal red local varieties was found to be less than 5 μg/g fresh wt., whereas the β-carotene content of various hybrids developed was found to vary from 20 to 38 μg/g fresh wt. The recommended daily requirement of vitamin A is 3,000 μg of β-carotene and it could be readily seen that a supplement in the diet of 150 g fresh wt. of the hybrids (2-3 fruits) will adequately meet the daily needs of vitamin A.

The detailed results of these studies will be communicated in the near future.

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1. Tomes, M. L., Quackenbush, F. W. and Kargl, T. E., *Botan. Gaz.*, 1958, **119**, 250.
2. Kargl, T. E., Quackenbush, F. W. and Tomes, M. L., *Proc. Amer. Soc. Hort. Sci.*, 1960, **75**, 574.
3. Jenkins, J. A. and Mackinney, G., *Genetics*, 1955, **40**, 715.
4. Williams, R. J. H., Britton, G. and Goodwin, T. W., *Biochem. J.*, 1967, **105**, 99.

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[illegible][1-3] reported a number of spray reagents for the detection of carbamates. In addition, other reagents[4-6] have also been used for the identification of carbamates. This laboratory is receiving considerable number of poisoning cases involving carbamate insecticides and we have tried Tollen's and Nessler's reagents, for their detection.

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TABLE I

The mineral contents in mammalian eye lenses

	Ca	Mg	Fe	Zn	Cu	Mn
	mg/100 g		←— μg/100 g —→			
Rat	3·5	0·48	580	—	—	—
Dog	5·6	4·1	68	1780·0	160·0	—
Cat	6·3	3·7	42	1535·0	148·0	—
Rabbit	3·8	0·51	670	18·3	4·1	8·1
Buffalo	4·9	0·96	106	216·0	216·0	27·3
Goat	4·3	1·02	119	246·0	290·0	20·1
Monkey	4·5	0·32	406	270·0	65·0	—

— not detected.

The values are calculated on the basis of wet weight of eye lenses and is the mean of four determinations each from a group consisting of 30–50 eye lenses.

selected for the present study as they belong to different orders of class mammalia.

The whole eye balls were obtained within two hours of the death of the animal. To minimize variation due to age, the eye balls from the adult animals were taken. They were carefully dissected and the lenses were freed from the capsule and the surrounding tissues on ice, blotted lightly to remove adhering moisture, dried in a vacuum desiccator to constant weight and ashed at 550° C. The acidified solution of ash was used for the estimation of calcium[1], magnesium[2], iron[3] copper[4], zinc[5] and manganese[6].

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1. Tomes, M. L., Quackenbush, F. W. and Kargl, T. E., *Botanical, Gaz.*, 1958, **119**, 250.
2. Kargl, T. E., Quackenbush, F. W. and Tomes, M. L., *Proc. Amer. Soc. Hort. Sci.*, 1960, **75**, 574.
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Baygon (2-Isopropaxy phenyl N-methyl carbamate)	0·34	..	0·34
Zineb (Zinc-ethylene-bisdithio carbamate)	0·98	..	0·98
Sodium diethyl dithio carbamate	0·54 (Brown)	..	0·54
Demecron (Phosphamidon) (Organo phosphate)]	0·44	0·44	Persists
Nuvan (Dimethyl Dichloro Vinyl Phosphate) (organophosphate)	0·38	0·38	Persists
Dipterex (organophosphate) [0, 0-dimethyl (2, 2, 2 trichloro-1-hydroxy ethyl) phosphonate)]	0·10	0·10	Persists
Dalf (Fenthion, Baytex)	0·63	0·63	Persists
(organo phosphorothionate)	0·38	..	..
	0·28	..	..
Dasanit (Fensulfothion)	0·34	0·34	Persists
	0·57	..	..
	0·92	..	..
Metasystox (oxydemetonmethyl)	0·10	..	...
(England) (Organo thiophosphate)	0·25	0·25	Persists

mercurous nitrate[7], which is commonly used for the detection of organo thiophosphates. These organophosphates react with Nessler's reagent and give black colour instantaneously whereas the carbamate insecticides give black colour with Nessler's reagent only after heating. This helps in differentiating the organophosphate insecticides from carbamate insecticides.

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---

1. Fishbein, L. and Fawkes, J., *J. Chromat.*, 1965, 19 (2), 364.
2. —, and Cavanaugh, M. A., *Ibid.*, 1965, 20, 283.
3. —, and Fawkes, J., *Ibid.*, 1965, 20 (3), 521.
4. Henkel, H. G., *Ibid.*, 1966, 21 (2), 346.
5. Post, A. P. and Stanley, C. W., *J. Agric. Fd. Chem.*, 1967, 15 (6), 1124.
6. Finochiaro, J. M. and Benson, W. R., *J.A.O.A.C.*, 1967, 50 (4), 888.
7. Kawala, G. B., Joglekar, V. D., Barve, V. P. and Mahal, H. S., *Sci. and Cult.*, 1972, 38, 373.

SIDA ACUTA— AN ADDITION TO THE HOST LIST OF *SCLEROTIUM ROLFSII* SACC.

Sclerotium rolfsii Sacc. [Corticium rolfsii (Sacc.) Curz.] is a tropical and subtropical pathogen whose intensity of attack is known to increase at high temperature under humid conditions. Gorakhpur with a humid subtropical climate offers an ideal locality for the occurrence of scleratial diseases of plants. Recently scleratial diseases of several plants have been reported[1-3], from the vicinity of Gorakhpur. This communication, however, deals with the report of sclerotial disease of *Sida actuta* Burm.

In August, 1975, the authors observed severe sclerotial rot of this plant at many places in the ıs of Gorakhpur. Under natural conditions, ɔptoms appeared as light brown water soaked s along the crown region of the plants which contract with the infested soil. The infected ıs were covered with radiating profuse myceal :. Later on, light brown, smooth surfaced, ɛ or elongate sclerotia measuring 0·5–1·0 mm ımeter developed on the mycelium. The ım gradually disappeared leaving the bare ia alone. The infected plants ultimately wilted

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, and —, *Ibid.*, (accepted), 1975.
, and —, *Indian Phytopath.*, (accepted), 1975.
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ımakrishnan, K. and Subramanian, C. V., *J. Madras Univ.*, 1956, 268, 327.
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ıkerji, K. G. and Juneja, R. C., *Fungi of India, Supplement to the List of Indian Fungi*, Emkay, Publications, Delhi, 1975, p. 224.

IUS MENONI, A NEW CYPRINID FISH
I BIHAR (INDIA) WITH A KEY TO THE INDIAN SPECIES OF *BARILIUS*

luction.—During the course of my studies on hwater fishes of Bihar, I came across a well d specimen belonging to the genus *Barilius* asuring 175·0 mm in total length, collected S. Biswas, 10–11–1968, from Gomatidhara, N. Bihar. This specimen on careful study, appeared to represent a hitherto unknown species. A detailed description of this new species along with a revised key of the genus *Barilius* to accommodate it is given in the present paper.

Description.—D. 9 (2/7), P. 14 (2/12), V. 9, A. 13, C. 23, L. 1. 62. Depth of body 3·25, length of head 3·88 in standard length. Width of head 2·00, height of head 1·33 in head length. Diameter of eye 4·5 in head length, 1·5 in snout length. Mouth anterior, oblique. having a deep cleft. Jaws compressed, the lower with a knob above the symphysis and an emargination to receive it in the upper jaw which is slightly longer. Interorbital space moderately convex, not flat. Length of snout 3·00, interorbital distance 3·27 in head length. Maxilla reaches below the middle of the eye. No barbel; even no trace of rudimentary ones. Dorsal fin commences midway between the anterior margin of the orbit and the base of the caudal fin and in advance of the anal fin. Pectoral reaches beyond the origin of the ventral fin. Auxilliary process prominent, extended beyond the bases of the pectoral and ventral fins. Predorsal scales 27. Distance between anterior origin of dorsal fin and tip of snout 1·81 in standard length. Length of pectoral 1·34 in length of head. Distance between anterior origins of pelvic and anal fins 2·30 between anterior origin of pelvic and base of caudal fin. Length of caudal peduncle 7·77 in standard length, 2·00 head length. Lateral line scales 62. Three and half rows of scales between lateral line and the base of ventral fin and thirteen and half rows of scales from lateral line to the base of dorsal fin. Colour in spirit, greyish on the back side, shiny golden yellow colour all over the body, dorsal and caudal fins slightly tinged with grey, 18 to 19 indistinct bluish vertical bars arranged in a single row on the dorsal side of the body.

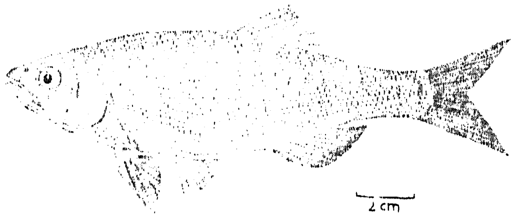

FIG. 1. Lateral view of the Holotype of *Barilius menoni* Sen (sp. nov).

Diagnosis.—No barbels. Lateral line scales 62. 18–19 indistinct bluish vertical colour bars arranged in a single row on the dorsal side of the body. Shiny golden yellow colour all over

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---

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5. Ramakrishnan, K. and Subramanian, C. V., *J. Madras Univ.*, 1956, 268, 327.
6. Tandon, R. N. and Chandra, S., *Supplement to the List of Indian Fungi*, Univ. Allahabad Studies, Allahabad, 1963, p. 246.
7. Mukerji, K. G. and Juneja, R. C., *Fungi of India, Supplement to the List of Indian Fungi*, Emkay, Publications, Delhi, 1975, p. 224.

BARILIUS MENONI, A NEW CYPRINID FISH FROM BIHAR (INDIA) WITH A KEY TO THE INDIAN SPECIES OF *BARILIUS*

Introduction.—During the course of my studies on the freshwater fishes of Bihar, I came across a well preserved specimen belonging to the genus *Barilius* and measuring 175·0 mm in total length, collected by Shri S. Biswas, 10–11–1968, from Gomatidhara, N. Bihar. This specimen on careful study, appeared to represent a hitherto unknown species. A detailed description of this new species along with a revised key of the genus *Barilius* to accommodate it is given in the present paper.

Description.—D. 9 (2/7), P. 14 (2/12), V. 9, A. 13, C. 23, L. l. 62. Depth of body 3·25, length of head 3·88 in standard length. Width of head 2·00, height of head 1·33 in head length. Diameter of eye 4·5 in head length, 1·5 in snout length. Mouth anterior, oblique. having a deep cleft. Jaws compressed, the lower with a knob above the symphysis and an emargination to receive it in the upper jaw which is slightly longer. Interorbital space moderately convex, not flat. Length of snout 3·00, interorbital distance 3·27 in head length. Maxilla reaches below the middle of the eye. No barbel ; even no trace of rudimentary ones. Dorsal fin commences midway between the anterior margin of the orbit and the base of the caudal fin and in advance of the anal fin. Pectoral reaches beyond the origin of the ventral fin. Auxilliary process prominent, extended beyond the bases of the pectoral and ventral fins. Predorsal scales 27. Distance between anterior origin of dorsal fin and tip of snout 1·81 in standard length. Length of pectoral 1·34 in length of head. Distance between anterior origins of pelvic and anal fins 2·30 between anterior origin of pelvic and base of caudal fin. Length of caudal peduncle 7·77 in standard length, 2·00 head length. Lateral line scales 62. Three and half rows of scales between lateral line and the base of ventral fin and thirteen and half rows of scales from lateral line to the base of dorsal fin. Colour in spirit, greyish on the back side, shiny golden yellow colour all over the body, dorsal and caudal fins slightly tinged with grey, 18 to 19 indistinct bluish vertical bars arranged in a single row on the dorsal side of the body.

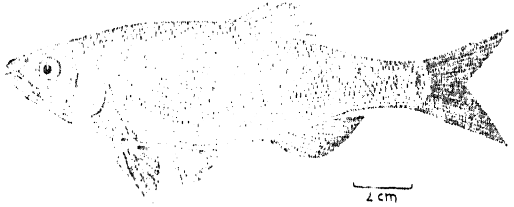

FIG. 1. Lateral view of the Holotype of *Barilius menoni* Sen (sp. nov).

Diagnosis.—No barbels. Lateral line scales 62. 18–19 indistinct bluish vertical colour bars arranged in a single row on the dorsal side of the body. Shiny golden yellow colour all over the body. Thirteen and half rows of scales between lateral line and anterior origin of dorsal fin and three and half rows of scales between lateral line and base of ventral fin.

Affinity.—*Barilius menoni,* in having no barbel, significantly differs from *B. radiolatus, B. shacra, B. bendelisis, B. dogarsinghi*[1], *B. barila, B. vagra,* in which there are either four or two barbels. *B. modestus* has been merged with *B. vagra*[3]. Without having no barbel, *B. menoni* resembles *B. evezardi, B. bakeri, B. canarensis, B. guttatus, B. tileo, B. bola* and *B. gatensis*; but markedly differs from all of them, in having 62 lateral line scales, 18–19 bluish indistinct vertical bars arranged in a single row on the dorsal side of the body, shiny golden yellow colour all over the body, thriteen and half rows of scales between lateral line and anterior origin of dorsal fin, which are present in none of them. All these significant differences distinguish it from all the so far known species of *Barilius.*

Holotype.—Reg. No. FF 750. Total length 175·0 mm., standard length 140·0 mm. Loc. Gomatidhara, Madhepura, District. Saharsa, N. Bihar. Coll. S. Biswas, dt. of coll. 10–11–1968. Deposited in the Zoological Survey of India, Calcutta.

Remarks.—The species is named after the eminent Ichthyologist, Dr. A. G. K. Menon, Dy. Director, Zoological Survey of India, who has kindly confirmed the identity of the new species.

	Revised key to the species of the genus	*Barilius*
1.	Barbel present	2
	Barbel absent	7
2.	Four barbels	3
	Two barbels	6
3.	Lateral line scales 56–70	4
	Lateral line scales 40–44	5
4.	Lateral line scales 56–62, No vertical bar	*B. radiolatus* (Günther)
	Lateral line scales 60–70, 12 vertical bars	*B. shacra* (Hamilton)
5.	Anal short with 9–11 rays, No vertical bar, each scale with a balck spot	*B. bendelisis* (Hamilton)
	Anal long with 13–17 ray, 10 vertical bars	*B. vagra* (Hamilton)
6.	Lateral line scales 38–39	*B. dogarsinghi* Hora (1921)
	Lateral line scales 43–46	*B. barila* Hamilton
7.	Body without vertical bars	8
	Body with vertical bars	13
8.	Body uniformly silvery	*B. evezardi* (Day)
	Body with rcws of spots	9
9.	With single row of large spots	*B. bakeri* (Day)
	With two rows of spots	10
10.	Lateral line scales 38–48	11
	Lateral line scales 70–94	12
11.	Lateral line scales 38	*B. canarensis* (Jerdon)
	Lateral line scales 44–48	*B. guttatus* (Day)
12.	Lateral line scales 70–75	*B. tileo* (Hamilton)
	Lateral line scales 88–95	*B. bola* (Hamilton)
13.	Lateral line scales 39–40, 15 vertical bars	*B. gatensis* (Valenciennes)
	Lateral line scales 62, 18–19 indistinct bluish vertical bars	*B. menoni* Sen

The author is thankful to Dr. S. Khera, Dy. Director-in-Charge, Z.S.I., for encouragement during the course of study, to Dr. B. Biswas, Dy. Director, for valuable suggestions, and to Dr. A. G. K. Menon, Dy. Director, for kindly confirming the identity of the species and for critically going through the manuscript.

Zoological Survey of India, T. K. SEN.
Calcutta-12, *July* 12, 1975.

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1. Hora, S. L., "Systematic description of the collection from the Manipur valley and the Naga Hills," *Rec. Indian Mus.,* 1921, 22, 191.
2. Misra, K. S., "An aid to the identification of the common commercial fishes of India and Pakistan," *Ibid.,* 1959, 57 (Parts 1–4), 1.
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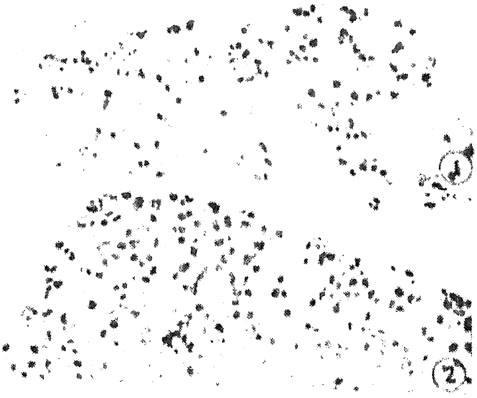
EFFECT OF CADMIUM CHLORIDE ON THE PERIPHERAL GONADOTROPHS IN THE HYPOPHYSIS OF RAT

CONFLICTING views are held by investigators with regard to the function of peripheral gonadotrophs in the rat[1-4]. So far it is not clear if the peripheral gonadotrophs secrete FSH or LH or both. The lacuna is due to the limited value of conventional histochemical tests applied by earlier workers. In recent times, however, the technique of immunohistochemistry is widely used for the recognition of various cell types in the hypophysis. In this con-test the peroxidase labelled antibody technique of Nakane and Pierce[5] is valuable for the detection of thyrotrophs and LH secreting cells in the rat hypophysis.

Male rats of the Wistar strain each weighing around 300 gms were used in the present study. They were kept in cages in the laboratory under seminatural conditions. To avoid overcrowding 5 rats were placed in each cage. One ml of aqueous solution of cadmium chloride (Cd: 0·04 M) was injected subcutaneously as recommended by Parizek[6]. 28 days after a single injection, rats were autopsied by decapitation, the pituitary glands were removed, weighed and fixed in Bouin's fluid. 6 microns thick sections of the hypophyses were stained immunohistochemically with peroxidase labelled goat antirabbit gamma globulin. 3,3'-diaminobenzidine was used as a substrate for the enzyme. A few sections of the hypophyses were also stained by PAS-orange G-alcian blue method of Herlant[7].

Administration of Cd resulted in a significant increase in the weight of the pituitary (11·44 mg ± 0·24) as compared with that of the controls (9·62 mg ± 0·18). Immunohistochemically, the peroxidase labelled antibody technique demonstrated both the thyrotrophs and LH secreting cells. Typically, the thyrotrophs are polygonal and vary in size. They are chiefly aggregated around the sinusoids. Cd does not seem to cause any change in either their size or number. LH secreting cells are rounded and occur on the dorsal and ventral regions of the hypophysis. The cells also vary in size. LH secreting cells are relatively few in the controls (Fig. 1). Cd administration induces hyperplasia and a slight hypertrophy in them (Fig. 2). On the contrary, based on conventional histochemical tests, Allanson and Deanesly[8] inferred that in cadmium injected rats the peripheral gonadotrophs are rounded and secrete only FSH, while the central gonadotrophs are typically oval and secrete LH.

Two types of PAS positive cells, viz., type I cells appearing round and colored brick red and type II cells which are oval and stained violet are observed in the hypophyses of controls as well as treated rats. Type I cells correspond to the LH secreting cells revealed by the immunohistochemical technique. Type II cells are similar to FSH secreting cells reported by Herlant[9]. Both type I and type II cells are more numerous peripherally on the dorsal and ventral aspects of the hypophyses in Cd treated animals than in the controls. The result of the present study corroborates the findings of Barnett *et al.*[1] that the peripheral gonadotrophs in the rat secrete both FSH and LH.



FIGS. 1-2. Fig. 1. Hypophysis of a control male rat stained with peroxidase labelled antibody showing both TSH cells and LH cells, × 400. Fig. 2. Hypophysis of a male rat that received CdCl. Note increase in the number of peripheral LH cells, × 400.

One of us (NHGD) is grateful to the Japan Society for the Promotion of Science, Tokyo, for the award of a Post-doctoral Fellowship during the tenure of which this work was carried out. We sincerely thank Prof. B. R. Seshachar, Indian Institute of Science, Bangalore, for critically going through the manuscript.

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* Post-graduate Department of Zoology, University of Mysore, Manasa Gangotri, Mysore 6, India.

1. Barnett, R. J., Ladman, A. J., McAllaster, N. J. and Siperstein, E. R., *Endocrinology*, 1956, 59, 398.
2. Hildebrand, J. E., Rennels, E. G. and Finerty, J. C., *Z. Zellforsch.*, 1957, 46, 400.
3. Rennels, E. G., *Ibid.*, 1957, 45, 464.
4. Purves, H. D. and Griesbach, W. E., *Endocrinology*, 1954, 55, 785.
5. Nakane, P. K. and Pierce, G. B. Jr., *J. Cell Biol.*, 1967, 33, 307.
6. Parizek, J., *J. Endocr.*, 1957, 15, 56.
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Zoological Survey of India, Calcutta-12, *July* 12, 1975.

T. K. Sen.

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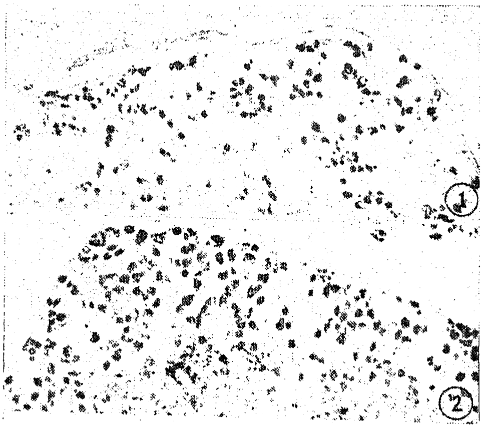

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AMINO ACID DIFFERENTIATION IN *OPHIOGLOSSUM* L. BY PAPER CHROMATOGRAPHY

THE genus *Ophioglossum* is a cosmopolitan plant belonging to Eusporangiate order of Filicales. The total number of species[1] cited for the genus is 45 out of which 10 species occur in India[2]. We are making morphogenetic studies on eight species collected from different localities in Central India and have found a few strains exhibiting serious abnormalities of tropophyll, spike and spores[3]. It is highly remarkable that other plants (growing at the same spot) of the same and different species do not possess such features, thus excluding any role of environmental factors. Since we have suggested that all these abnormalities may be correlated with certain genetic phenomenon, we thought of investigating certain differences in normal and abnormal plants of different species. As amino acids are influenced by the genes, the identification of these acids may lead to the differentiation of the species. This paper is a part of the study dealing with separation of amino acids contained in the general pools of different organs in mature plants.

Plants were collected from Gwalior (Kanhar Jhir, Devkho and Tighra), Shivpuri and Nainital and the amino acid separation has been done on *O. costatum* R. Br., *O. nudicaule* L., *O. petiolatum* Hook., *O. lusitanicum* L., *O. vulgatum* L., *O. gramineum* var. *gramineum* Willd. The plants were thoroughly washed with water and separated into leaf, spike and rhizome bearing roots. Each material was extracted with few cc of 10% isopropylalcohol[5,6]. The amino acids were separated by paper chromatography employing stripe (5–6 cm wide) of Whatman No. 1 filterpaper. A mixture of N-butanol, acetic acid, water (12 : 3 : 5) was employed as the solvent for the chromatography. After 12–16 h. of irrigation, the paper was dried then sprayed with a solution of ninhydrin and allowed to dry. The paper was then heated for 5 minutes at about 80° C to develop the ninhydrin colour and the zones so produced were outlined with a pencil. The R_F values were calculated from these chromatograms and compared with the R_F value of known aminoacids obtaintd under identical conditions and the aminoacids in the unknown solutions were identified.

We have consistently found that cysteine and ornithine are exclusively present in the rhizome of all the species investigated but not in the tropophyll, petiole and spike. This may suggest the probable role of these aminoacids in vegetative propagation of plants. Eight other aminoacids were detected in the genus *Ophioglossum* (Table I). Another

TABLE I

Comparative chart of Amino acid separation in Ophioglossum *by paper chromatography* (*single dimension*)

Sl. No.	Amino-acids	*O. costatum*			*O. vulgatum*			*O. gramineum*			*O. lusitanicum*			*O. nudicaule*			*O. petiolatum*		
		R.	S.	L.	R.	S.	L.	R.	S.	L.	R.	S.	L.	R.	S.	L.	R.	S.	L.
1.	Cysteine	+	—	—	+	—	—	+	—	—	+	—	—	+	—	—	+	—	—
2.	Ornithine	+	—	—	+	—	—	+	—	—	+	—	—	+	—	—	+	—	—
3.	Lysine	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
4.	Arginine	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
5.	Threonine	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
6.	Alanine	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
7.	Proline	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
8.	Tyrosine	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
9.	Valine	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
10.	Norleucine	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
11.	Unknown	+	+	+	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
12.	Unknown	+	+	+	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—

Abbreviations used

R. = Rhizome with roots, S. = Spike, L. = Leaf, + = Aminoacid Present, and — = Aminoacid Absent.

important observation concerns with the two spots of the unknown aminoacids below norleucine in the extracts of leaf, spike and rhizome of the plants (*see* Table I) comparable with *O. costatum*. These plants however differ with *O. costatum* in the shape and sizes of tropophyll and often bear spikes which abort before reaching maturity. A quantitative analysis of aminoacids in the organs of the species *Ophioglossum* can be of great value in the identification.

We are indebted to Prof. M. S. Agarkar, for his help with plant collections. Our thanks are due to Dr. R. P. Bhatnagar, Department of Chemistry and Dr. R. Mathur, Department of Zoology, Jiwaji University, for supervision of the work. One of us (SK) is grateful to the U.G.C. (India), for the Junior Research Fellowship.

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October 31, 1974.

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1. Sporne, K. R., *The Morphology of Pteridophytes*, Hutchinson and Co, Ltd., 1970, p. 135.
2. Panigrahi, G. and Dixit, R. D., *Proc. Nat. Inst. Sci. India*, 1969, **35 B**, 3.
3. Goswami, H. K. and Khandelwal, S., *Brit. Fern. Gaz.*, 1973, **10** (6), 311.
4. Hunter, G. E., *Amer. J. Bot.*, 1967, **54** (4), 473.
5. Weissberger, A., *Technique of Organic Chemistry*, Vol. V, Cassid, H. G., *Adsorption and Chromatography*, Interscience Publisher, Inc., New York, 1951, p. 307.
6. Zweig, G. and Whitaker, J. R., *Paper Chromatography and Electrophoresis*, Vol. II, Sherma, J. and Zweig, G. *Paper Chromatography*, Academic Press, Inc., (London), Ltd., 1971, p. 720.

A NEW SPECIES OF *SPORIDESMIUM*

DURING a survey of fungi from Mount Abu, Rajasthan, the authors observed thick and extensive, black fungal growth on leaf bases of *Phoenix sylvestris* attached to the tree trunks. The fungus was found to be a new species of *Spirodesmium*. The present fungus is very close to *Spirodesmium fragilissimum* (Berk. and Curt.) M. B. Ellis[1] but differs in having very long conidiophores and conidia. In case of *S. fragilissimum* the proximal half of the conidial wall is verrucose, whereas in the present fungus the entire wall may be verrucose in most of the conidia. Due to these distinct morphological characters this fungus is being described as a new species.

Spirodesmium longissimum sp. nov. Panwar and Chouhan.

Coloniae effusae, atrae, mycelium in parte superficiale sed pro parte in substrato immersum, cum ramis, septatum, olivaceum-brunneum, levi, pariete, 3–5·5 μ crasse hyphae. Conidiophora a latere super hyphis surgentia in turmis, perphyrea, erecta, simplicia, recta vel leviter flexa, paries levis raro verrucosus, 8–38 septata, septa crica 6·5–32·0 μ seorsum, cum usque ad 3 successivae proliferationes, 220–660 μ longae, 5·4–6·8 μ crassae. Conidia singulatim formata apud apicem, conidiophorum quae post seperationem primi conidii, prolifert per cicatricem, et fit, alter conidium apud spicem proliferationis, rectum vel leviter flexum, obclavatum, truncatum vel raro conica truncatum apud basim, dilutum olivaceum brunneum ad olivaceum brunneum, distale extremum subhyalinum, paries, vel omino verrucosus vel dimedium proximale verrucosus, rectum vel contractum, ad septa transversa, 7–39 septata, 50–311 μ longa, 5·5–8 μ crassa, in parte latissima, contracta ad 2·8–5·0 μ ad apicem, 4·0–5·0 μ lata ad basim, septa circiter 5·4–12 μ seorsum [Fig. 1 (*a*), 1 (*b*)].

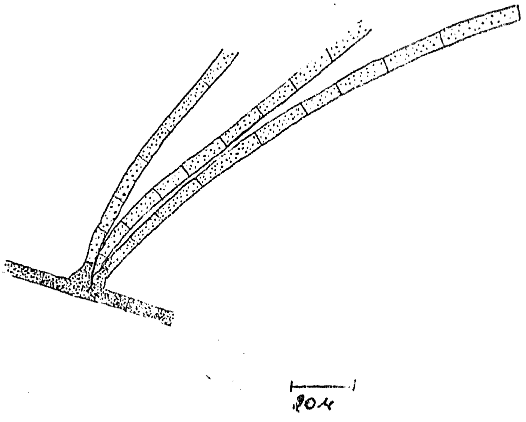

FIG. 1 (*a*) Conidiophores in a group.

Basi arido folio phoenix sylvestris, Mt. Abu, August, 1973. Specimen depositum apud C.M.I., Kew, Herb. IMI 180041 typus. Coll. No. J.U.M.L. 321.

Colonies effused, black;; mycelium partly superficial but mostly immersed in the substratum, composed of branched, septate, olivaceous-brown, smooth walled, 3–5·5 μ thick hyphae. Conidiophores arising laterally on the hyphae in groups, dark reddish-brown, erect, simple, straight or slightly flexuous, wall smooth, rarely verrucose, 8–38 septate, septa averaging 6·5–32·0 μ apart, with upto 3 successive proliferations, 220–660 μ

long, 5·4–6·8 μ thick. Conidia formed singly at the apex of the conidiophore which after the first conidium has fallen sometimes proliferates through the scar and forms another conidium at the apex of the proliferation, straight or slightly flexuous, obclavate, truncate or rarely conico-truncate at the base, pale olivaceous-brown to olivaceous-brown, distal end sub-hyaline, wall either completely verrucose or proximal half verrucose, straight or constricted at the transverse septa, 7–39 septate, 50–311 μ long, 5·5–8 μ thick in widest part, tapering to 2·8–5·0 μ near the apex, 4·0–5·0 μ wide at the base, septa averaging 5·4–12 μ apart [Fig. 1 (*a*), 1 (*b*)].

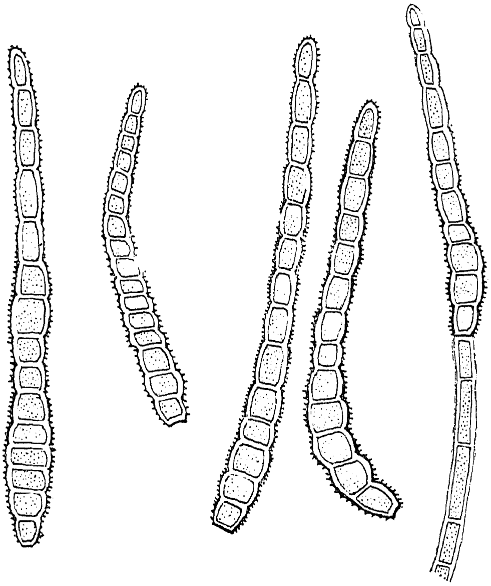

FIG. 1 (*b*) Conidia.

On drying leaf bases of Phoenix sylvestris, Mt. Abu, August, 1973. Specimen deposited with C.M.I., Kew, Herb. IMI 180041 type. Coll. No. J.U.M.L. 321.

We are grateful to Dr. M. B. Ellis, for the identification of the fungus. Thanks are due to Prof. H. C. Arya, for providing laboratory facilities and to Rev. Father William Barracos, for the Latin translation.

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Department of Botany,
University of Jodhpur,
Jodhpur, India, *November* 25, 1974.
K. S. PANWAR.
J. S. CHOUHAN.

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1. Ellis, M. B., *Mycol. Paper*, 1958, 70, 55.

COLLETOTRICHUM ACUTATUM—A NEW SEED BORNE PATHOGEN OF *ZINNIA*

DURING seed health testing of seeds of *Zinnia elegans* and *Z. haageans* an unidentified species of *Colletotrichum* was encountered by Neergaard, 1966 at the Plant Protection Service, Denmark. In 1972 a seed sample of *Zinnia* recieved from Italy, when tested on blotter method for their seed borne fungi, was found 5% infected by *Colletotrichum* species. On comparison with other known species of *Colletotrichum* this isolate was identified as *Colletotrichum acutatum* Simmonds.

On seeds the fungus forms acervuli, mostly single, rarely in groups, with numerous setae. Setae are brownish black, minute, smaller than spore mass. Spore mass bright orange. Mycelium rare or absent (Fig. 1 A).

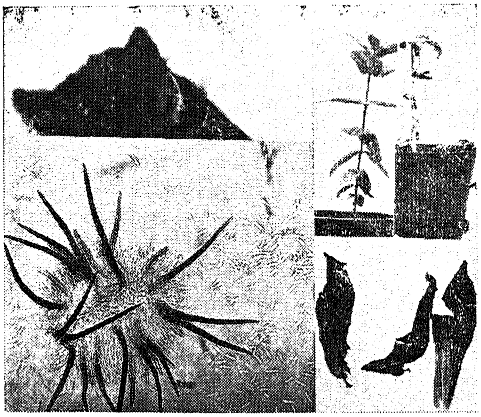

FIG. 1. *Colletotrichum zinniae* sp. nov. on *Zinnia elegans*. (A) acervuli on seeds, × 175; (B) structure of acervulus showing conidia, conidiophores, and setae × 500; (C) Healthy seedling; (D) infected seedling; (E) Shrivelled, deformed and distorted leaves.

On agar medium colonies slow growing, restricted, effused, 15 mm in diameter within a week, pink orange to bulff orange, margine smooth. Reverse pink orange. Acervuli formed in zones, minute, lyso-orange. Setae trichiformis, blackish-brown.

Conidia hyaline, one celled, cylindrical, fusoid, straight, pointed at both ends, 6·0–12·0 × 1·5–2·5 μ. Setae numerous, trichiform, brownish black 1–4 septate, 30·0–90·0 × 3·0–6·0 μ (Fig. 1 B).

In order to test the pathogenicity two to three weeks old seedlings of *Z. elegans* were inoculated by spraying with spore suspension of *C. acutatum* under glass house condition. The seedlings started showing symptoms within four days. The leaves became shrivelled, deformed, severe necrosis developed on the stem and seedlings were killed within a week (Fig. 1 D–E).

This fungus has been reported for the first time on this host. A number of other hosts of this pathogen has been reported by Simmonds (1965) after isolating it from *Carica papaya*. Since this pathogen is seed borne, it may easily be introduced into new areas.

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Indian Agricultural Research Institute,
New Delhi 110 012, *March* 21, 1975.
D. D. KULSHRESTHA.

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1. Neergaard, Paul, *Arsberetning Vedorende, Froptologisk Kontrol*, 1. June 1964, 31 May, 1965, Statens Plantetilsyn, Kobenhavn, 1966, 17.
2. Simmonds, J. H., *Qd. J. Agric. Sci.*, 1965, 22, 437.

OPHIOGLOSSUM VULGATUM LINN.—A NEW HOST OF *CURVULARIA LUNATA* (WAL.) BOED

GORAKHPUR District being fairly moist exhibits a luxuriant growth and display of ferns and fernallies[1]. This situation offers possibilities of incidence of interesting fungal parasites of these plants. But parasitic fungal flora of Pteridophytes of this region has not yet been explored, barring a few reports[2]. During routine survey of fungi causing plant diseases in this locality, authors came across a large number of plants of *Ophioglossum vulgatum* showing leaf blight symptoms in December, 1974.

The early infections manifested in the form of small water-soaked circular spots particularly in the middle of the fronds. Later on the spots coaliced forming large pale yellow green patches due to the death of the infected tissues. Examination of the field specimen of the infected fronds showed the presence of *Curvularia* species. Repeated isolations made on Czapek's solution agar also yielded this fungal species. The fungus was characterized by spreading, subfloccose, dark olive-gray colonies; stout, septate, brownish, thick walled, unbranched, 72·0–100 × 2·7–4·5 μ conidiophores; alternately arranged, three septate, curved, brown, 25·2–34·2 × 9·0–11·4 μ conidia (Fig. 1).

The features mentioned above agree pretty well with *Curvularia lunata* in its essential characters except that the conidiophores are slightly smaller and conidia are larger in their measurements. The herbarium specimen is maintained as GPU Herb. No. 112, S. Singh. It is also being sent to the Indian type culture collection, I.A.R.I., New Delhi and Commonwealth Mycological Institute, Kew, England. This appears to be a new host record for *Curvularia lunata* (Walk.) Boed. from India.

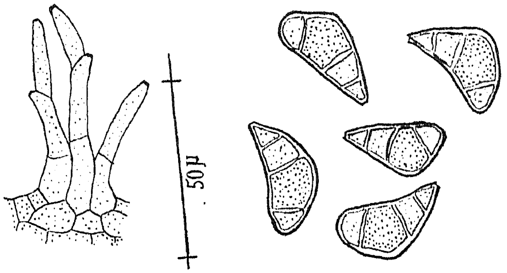


FIG. 1

Authors wish to express their grateful thanks to Prof. K. S. Bhargava for his keen interest and for providing facilities.

Department of Botany,
University of Gorakhpur,
Gorakhpur (U.P.), India,
January 9, 1975.
KAMAL.
S. SINGH.

---

1. Sharma, D. N., Tripathi, S. M. and Srivastava, A. K., *Indian Forester*, 1969, 95, 526.
2. Bhargava, K. S. and Misra, P. C., *Sydowia, Annales Mycologici. Ser. II*, 1961, 15, 231.

ON THE OCCURRENCE OF STOMATA ON THE INNER OVARY WALL OF *BELAMCANDA CHINENSIS* DC.

REMARKABLY little is known about the inner ovary wall (epidermis) despite frequent studies[1,2], in recent years on stomatal ontogeny and organography and there is no work in which the entire stomatiferous area of a plant species has been examined. In this note the occurrence of stomata on the inner ovary wall of *Belamcanda chinensis* DC (Iridaceae) and their ontogeny is described which are not recorded in the literature.

Epidermal peels from the inner and outer walls of young and old flowers and mature leaves of *B. chinensis* were stained in aceto-carmine, washed and mounted in 50% glycerine for observations.

In Fig. 1, longisection of inferior ovary of *B. chinensis* and the area on which stomata have been seen are shown. The family Iridaceae is known to have Allium type of stomata[5] which are devoid of subsidiary cells but have neighbouring cells without derivatives[6]. Mature stomata are evenly distributed all along the inner and outer walls in linear rows parallel to the axis of the plant body. The stomata of the inner ovary wall are in the level of the epidermal cells while those of

Table I

Comparison of stomatal characteristics of ovary and leaf in B. chinensis

		Stomatal frequency	Size in microns	Number of neighbouring cells	Size
Ovary	inner wall	3/mm^2	21·1 × 18·0	6 to 9	large
	outer wall	6/mm^2	23·1 × 23·8	8 to 12	small
Leaf (isobilateral)		10/mm^2	60·7 × 43·4	4 to 5	very large

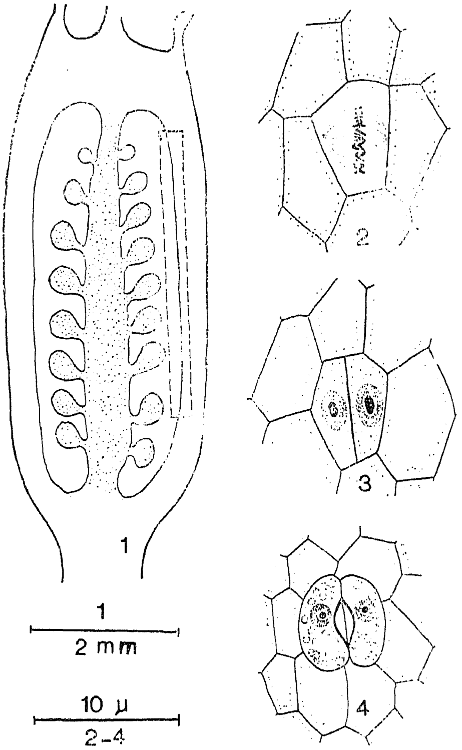
* mean of 25 readings.

the outer are considerably sunken below the level of the epidermis. In transverse section stomata from the inner ovary wall show normal guard cells with thickening wall and faint cuticular ledges. Table I summarises the stomatal characteristics of ovary walls and leaves. The values of stomatal size and frequency are highest for the leaf and diminish as we go to outer and inner walls. Further, the size and the number of neighbouring cells is different in all the three cases. Epidermal wall thickening on the inner wall is less conspicuous than the outer. The stomata of the inner wall appear to be functional with several chloroplasts and the pore is likewise conspicuous (Fig. 2) indicating photosynthetic activity.

What role the inner wall stomata play in transpiration remains to be elucidated.

In their ontogeny, inner wall stomata differenciate rather early. A protoderm cell itself becomes a meristemoid which can be recognised by its large size and dense contents. Meristemoids are oriented in parallel rows. The meristemoid becomes spherical and directly functions as guard mother cell, and divides (by a wall) to give rise to two equal guard cells (Figs. 3 and 4). The ontogeny of outer wall stomata and leaf is similar. In all the cases the neighbouring cells show no division and become direct contact cells.

The present observations of the occurrence of stomata on the inner ovary wall in *B. chinensis* are significant in that these have not been recorded earlier and further they shed basic information on the morphology of the carpel. The inner wall of the ovary and the placentae are considered carpellary structures[3,4]. The presence of stomata on the inner ovary wall lends support to the foliar nature of the carpel which is well known. Similar observations have been made on *Eleutherine* sp. (Iridaceae) and other monocotyledons and a correlation seems to exist between the presence of stomata on the inner ovary wall and the inferior ovary (Bir Bahadur, unpublished).



Figs. 1–4. Fig. 1. Diagrammatic longisection of inferior ovary of *B. chinensis*, the inner ovary wall (boxed) on which stomata have been seen. Fig. 2. Mature stomata from the inner ovary wall. Figs. 3 and 4. Stages in the ontogeny of inner wall stomata.

The author is grateful to Prof. U. B. S. Swami, for facilities and encouragement and to Dr. M. R.

Pa[illegible] Marathwada University for useful discussion.

Department of Botany, Bir Bahadur.
Post Graduate Centre,
Osmania University,
Warangal 506 009 (A.P.), [illegible] 1975.

1. Bir Bahadur, Rangopal, [illegible] and Ramayya, N., *Bot. J. Linn. Soc. (Lond.)*, 1971, 64, 295.
2. Gupta, S. C., Paliwal, G. S. and Gupta, M., *Ann. Bot. (Lond.)*, 1965, 29, 615.
3. Pa[illegible] N., *Phytomorphology*, 1952 a, 2, 122.
4. —, *Bot. Rev.*, 1952 b, 18, 604.
5. Stebbins, G. L. and Khush, G. S., *Am. J. Bot.*, 1961, 48, 51.
6. Tomlinson, P. B., *Taxon*, 1974, 23, 109.

OCCURRENCE OF TYPES WITH CHARACTERS OF *ORYZA GLABERRIMA* IN ASSAM RICE COLLECTION

Character association leading to subspecies and species formation as a concept, is known in rice. Characteristic features of the *japonica* (considered a subspecies of *Oryza sativa*) have been mentioned by Nagai (1959). Swaminathan *et al.* (1968) had suggested that disruptive selection leading to constellation of characters was a force in subspecies development in *O. sativa*. In the varieties belonging to the African cultivated species, the common features include short and truncate ligule, unbranched panicle habit, glabrous hull and leaf in general, (with exceptions), red kernel and absence of perennating habit (Seetharaman, 1962). While a few of these features are also possessed by many of the *indica* varieties, forms with short and truncate ligule and unbranched panicle were never observed in *indica* varieties. The occurrence of such varieties is now reported.

The Assam Rice Collection (ARC) consisting of 7000 varieties collected from Assam and other areas in N.E. India by Swaminathan and his colleagues was transferred for maintenance to the Central Rice Research Institute in 1972. During the course of observation, six entries had some of the distinguishing features of *O. glaberrima* (Table I).

The varieties resemble the *glaberrimas* in (a) having short truncate ligule, (b) the absence of secondary branches in the panicle (with rare exceptions) and (c) having glabrous hull with red kernel. Red kernel and glabrous hull are also to be seen in the varieties belonging to *O. sativa*.

Occurrence of short ligule and panicles without (or rare) secondary branches in cultivars from India had not been reported earlier and is of significance. In mutation and hybridization studies carried out by Ratho (unpublished thesis) in the two species (*O. sativa* and *O. glaberrima*), it was reported that these two characters remained together. It is an example of parallel evolution through mutation in the forms belonging to the two cultivated species. It may, therefore, be inferred that the genes for panicle habit and nature of ligule are separate but are situated close together. It is a case of close linkage rather than pleiotropy.

TABLE I

Information on the varieties

ARC No.	Height (cm)	Spikelets per panicle (no.)
6136	84	40
6137	[illegible]	[illegible]
6189	[illegible]	34
6191	53	20
6193	92	38
6200	81	29
Oryza glaberrima	95	60

The authors are thankful to Dr. S. Y. Padmanabhan, Director, for his suggestions.

Genetics Division, R. SEETHARAMAN.
Central Rice Research D. P. GHOSAL.
Institute,
Cuttack 753 006, Orissa,
August 8, 1975.

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2. Seetharaman, R., *Sci. and Cult.*, 1962, 28, 286.
3. Siddiq, E. A. and Swaminathan, M. S., *Tech. Bull.* 86 (*Rice Breeding with Induced Mutation* II), IAEA, Vienna, 1968, p. 25.
4. Ratho, S. N., "Cytogenetical studies in the African cultivated rice, *O. glaberrima*, steud with special reference to the exploitation of this germ plasm," Unpub., *Ph.D. Thesis*, Utkal Univ.

A NOTE ON THE EMBRYOLOGY OF *LIPOCARPHA SPHACELATA* KUNTH (CYPERACEAE)

The earlier embryological investigations of the genus *Lipocarpha* have been concerned only with the development of embryo[3-6]. *L. chinensis* (Osb.) Kern (*L. argentea* R. Br.) and *L. sphacelata* Kunth (*L. triceps* Nees) have been recorded from South India[1]. The present investigation deals with the development of the male and female gametophytes, endosperm and embryo in *L. sphacelata*.

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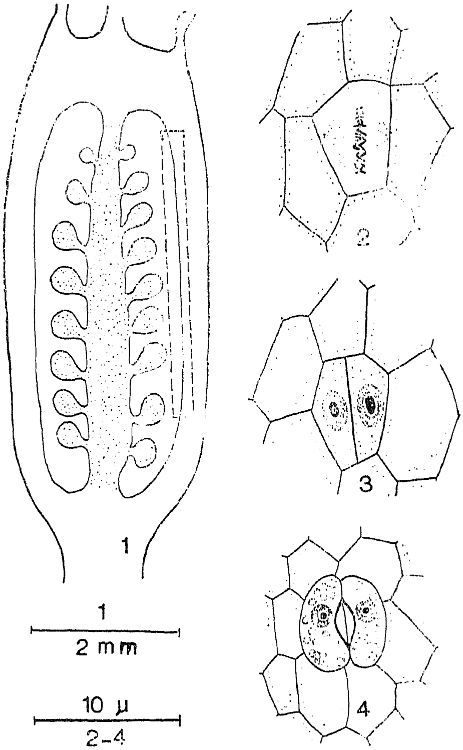

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---

1. Bir Bahadur, Rajagopal, T. and Ramayya, N., *Bot. J. Linn. Soc.* (*Lond.*)., 1971, 64, 295.
2. Gupta, S. C., Paliwal, G. S. and Gupta, M., *Ann. Bot.* (*Lond.*)., 1965, 29, 645.
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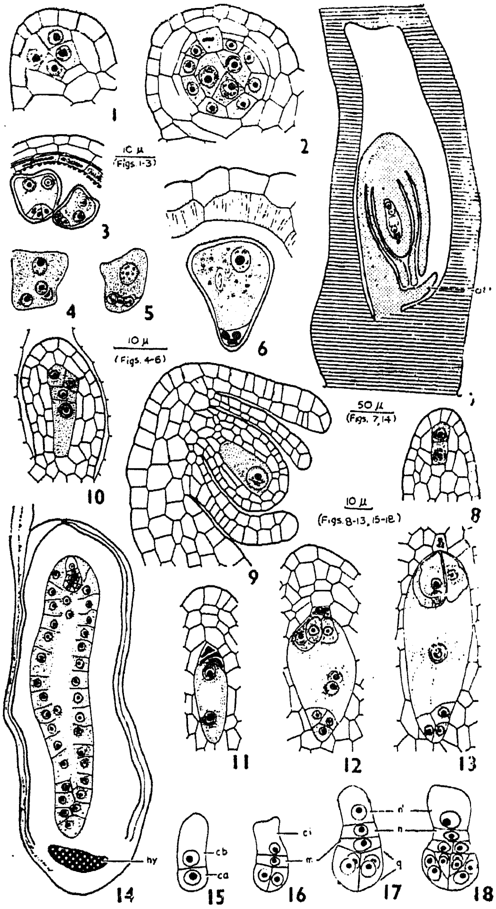
Lipocarpha sphacelata is a glabrous herb with tufted stems bearing several dark-purple spikelets grouped in terminal bracteate "heads". The flowers are bisexual each with 2 hypogynous scales, within which there are 2 stamens and a tricarpellary gynoecium. The ovary is unilocular with a basal ovule (Fig. 7).

One or two hypodermal archesporial cells are differentiated in each lobe of the 4-lobed young anther as seen in transection. They divide periclinally forming an inner primary sporogenous layer and an outer primary parietal layer (Fig. 1). The latter by further periclinal and anticlinal divisions forms an endothecium, a middle layer and a part of the tapetum (Fig. 2). The remaining part of the tapetum is derived from the cells of the connective adjacent to the sporogenous cells. Thus the tapetum is of dual origin. As the anther development proceeds, the middle layer degenerates and the cells of the endothecium develop thickenings (Figs. 3, 6). The tapetum is of the glandular type and its cells remain uninucleate throughout, and later degenerate. Ubisch bodies are observed along the inner side of the tapetum (Fig. 3).

The primary sporogenous cells divide and the products of the last division form the microspore mother cells. Each microspore mother cell undergoes meiosis and forms 4 nuclei of which 3 occupy the wallward position while the fourth remains in the center (Fig. 4). The cytokinesis is of the simultaneous type resulting in the formation of a tetrad (Fig. 5). The latter consists of 3 small non-functional microspores which later degenerate *in situ* and a large functional microspore which develops into a pollen grain. The pollen grains have a thick exine and a thin intine, and are 3-celled when shed (Fig. 6).

The ovule is anatropous, bitegminal and crassinucellar. Some of the funicular cells elongate and later cover the outer opening of the micropyle to from an obturator (Fig. 7). After fertilization, a group of cells at the chalazal region become thick-walled and form a hypostase (Fig. 14).

A single archesporial cell is differentiated in the ovule primordium and divides forming an outer primary parietal cell and an inner sporogenous cell (Fig. 8). The former divides and forms a 3- or 4-layered parietal tissue. The sporogenous cell functions as the megaspore mother cell (Fig. 9). The megaspore mother cell undergoes meiosis and usually forms a linear tetrad of megaspores; occasionally a T-shaped tetrad is observed (Fig. 10).



FIGS. 1–18. Fig. 1. T.s. a portion of young anther showing the primary parietal cells and the primary sporogenous cells (stippled). Fig. 2. T.s. a microsporangium showing the anther wall and the sporogenous tissue. Fig. 3. T.s. a portion of microsporangium showing degenerating tapetal cells and 2-celled pollen grains. Fig. 4. A microspore mother cell with free microspore nuclei. Fig. 5. A microspore tetrad. Fig. 6. T.s. a portion of microsporangium showing thickenings in the endothecium and 3-celled pollen grain; note the remnants of non-functional microspores. Fig. 7. L.s. ovary with an ovule showing 4-nucleate embryo sac and an obturator. Fig. 8. L.s. ovule primordium showing the primary parietal cell and the sporogenous cell (stippled). Fig. 9. L.s. ovule with megaspore mother cell. Fig. 10. L.s. nucellus with a T-shaped megaspore tetrad. Fig. 11. 2-nucleate embryo sac. Fig. 12. 8-nucleate embryo sac. Fig. 13. Mature embryo sac. Fig. 14. L.s. young seed showing wall formation in the endosperm. Fig. 15. 2-celled proembryo. Fig. 16. Proembryonal tetrad. Figs. 17 and 18. Quadrant and octant stages of the embryo. (*ca* and *cb*, terminal and basal cells of the 2-celled proembryo; *ci*, lower daughter cell of *cb*; *hy*, hypostase; *m*, middle cell of the proembryonal tetrad; *n* and *n'*, upper and lower daughter cells of *ci*; *obt*, obturator; *q*, quadrant).

The chalazal megaspore functions while the other 3 degenerate. The nucleus of the functional megaspore undergoes 3 successive divisions and forms 2-, 4- and 8-nucleate embryo sac of the Polygonum type[4] (Figs. 7, 11, 12). The organized embryo sac has an egg apparatus, 2 polar nuclei and 3 antipodal cells (Fig. 12). The polar nuclei fuse in the center of the embryo sac to form the secondary nucleus (Fig. 13).

Fertilization is porogamous. Syngamy and triple fusion take place resulting in the formation of the zygote and the primary endosperm nucleus respectively. The first division of the primary endosperm nucleus is not followed by wall formation. Free-nuclear divisions take place in the endosperm until about 120 nuclei are produced. Then centripetal wall formation sets in and ultimately the endosperm becomes cellular (Fig. 14). The zygote divides transversely forming a terminal cell *ca* and a basal cell *cb* (Fig. 15). The former undergoes 2 vertical divisions one at right angles to the other and forms a quadrant *q* (Figs. 16, 17). The cell *cb* divides transversely forming 2 superposed cells *m* and *ci*. The cells of quadrant divide by oblique walls delimiting the protoderm from an inner group of cells (Fig. 18). The embryo development conforms to the Juncus variation of the Onagrad type[2] and the disposition of different regions in the mature embryo corresponds to the Cyperus type[5].

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---

1. Fischer, C. E. C., *Flora of the Presidency of Madras*, (Reprint), B.S.I., Calcutta, 1957, 3, 1158.
2. Johansen, D. A., *Plant Embryology*, Waltham, Mass., 1950, p. 116.
3. Juguet, M., *Adansonia*, Ser. 2, 1970, 10, 271.
4. Maheshwari, P., *An Introduction to the Embryology of Angiosperms*, McGraw-Hill, New York, 1950, p. 87.
5. Shah, C. K., *Phytomorphology*, 1965, **15**, 1.
6. Van der Veken, P., *Bull. Jard. Bot. Etat. Belg.*, 1965, 35, 285.

A NOTE ON THE INDUCED VARIATIONS IN CHILLIES

THE crop improvement work done hitherto was by selection and hybridisation in chillies. Success achieved in evolving improved varieties in other crop plants in recent years[1], prompted the use of induced mutagenesis in the genetic improvement of chillies. Physical and chemical mutagens, namely, gamma rays, X-rays and ethyl methane sulphonate (EMS) were used on two local varieties of chillies, mundu and sattur samba.

Gamma irradiation was done using 2000 Curie ^{60}Co gamma cell. X-irradiation was done from Philips C. T. apparatus operated at 50 kV without filter at a distance of 4 cm from the source, the dose rate being 500 R per minute. The doses of gamma rays and X-rays employed were 10 to 60 kR with a stepwise interval of 10 kR. The seeds pre-soaked in distilled water for 6 hours were treated with six different concentrations of aqueous EMS (20 to 70 mM) for 4 hours. For each dose 100 seeds were treated and M_1 population was raised. In the present note a few of the morphological variations noticed in M_1 in both the varieties are described.

The untreated seedlings of mundu show apical dominance and alternate phyllotaxy. One of the seedlings at 10 kR of X irradiated M_1 population showed supression of apical dominance and branching at the early seedling stage while the other showed three cotyledonary leaves. One of the M_1 plants of mundu at 40 mM of EMS treatment showed the occurrence of aerial roots in the basal portions of the stems which were not observed in the control. The flowers borne on the untreated plants have five petals. The number of stamens in the flowers are equal to those of petals. However, among the 65 M_1 plants, treated with gamma rays (30 kR) five showed variations in the number of petals and anthers ranging from 4 to 7.

The untreated plants of sattur samba possess 4–6 primary branches on the mainstem. The mutant identified in 50 mM of EMS treated M_1 plants exhibited uniculm with the supression of primary branches. In the six out of 59 M_1 plants of sattur samba treated with 40 mM of EMS, some of the flowers had larger number of petals and with more than one ovary although the majority showed normal flowers. The fruits developed from these abnormal flowers showed variations in size. Rarely, on the same pedicel 1 or 2 extremely small fruits with varying size could be seen. The phenotypic changes noticed in the M_1 showed the effect of mutagens on chillies.

The writer is grateful to Dr. C. V. Govindaswamy, Dean, Dr. P. Chandrasekharan, Professor of Botany, for providing necessary facilities.

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---

1. Sigurbjornsson, B., "Breeding with natural and induced variability," *Induced Mutations and Plant Improvement*, STI/PUB/297, IAEA, Vienna, 1972, p. 3.

STUDIES ON STORAGE DISEASES OF FRUITS AND VEGETABLES

IV. Control of Guava Fruit Rot Caused by *Pestalotia psidii* and *Colletotrichum gloeosporioides* with Aretan

ARETAN, an ethoxyethyl mercuric chloride (6% Hg) compound is cheaper than many fungicides and antibiotics which have been evaluated for checking *Pestalotia psidii* Pat. and *Colletotrichum gloeosporioides* Penz. rots of guava. It has given very promising results with a number of diseases caused by diverse pathogens[1-3]. The efficacy of this fungicide in checking these two rots of guava was evaluated and the results have been presented in this note.

Preliminary *in vitro* study was carried out employing food poison technique. When the organisms were grown on Asthana and Hawker's medium 'A' supplemented with this compound, it was noted that 1 and 50 ppm of the compound was sufficient to completely check the growth of *P. psidii* and *C. gloeosporioides* respectively. These effective concentrations of the compound were then evaluated for their efficacy in checking the two diseases on artificially infested guava fruits (variety 'Safeda'). Both pre- and post-inoculation treatments were given by dipping the fruits in effective concentrations of the compound for 2 minutes. The treated fruits were incubated for 8 days at 28° C in glass chambers.

No sign of rotting was observed on the fruits treated with fungicide either before inoculation or after inoculation. Fruits infected with *P. psidii* and given pre-inoculation treatment became slightly soft but the softness could not be attributed to the activity of pathogen because its presence could not be detected in the fruits. As both the rots could be successfully checked by a low concentration of aretan, the compound may be recommended for the control of these diseases.

Authors are grateful to Prof D. D. Pant, for providing laboratory facilities.

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Allahabad 211 002, *August* 23, 1975.

---

1. Hide, G. A., Hirst, J. M. and Mundy, E. J., *Ann. Appl. Biol.*, 1969, **64**, 265.
2. Patil-Kulkarni, B. C. and Aberdeen, J. E. C., *Aust. J. Exp. Agric. Anim. Husb.*, 1969, **9**, 457.
3. Sharma, S. L. and Sohi, H. S., *Indian Potato, J.*, 1965, **7**, 12.

EFFECTIVENESS OF KNOWN *Sr* GENES FOR SEEDLING RESISTANCE AGAINST INDIAN STEM RUST RACES IN WHEAT

WHEAT rusts pose a serious threat to wheat production in India through the high yielding varieties programme, during the last decade. The problem has attracted particular attention with *Kalyan Sona,* one of the most important contributors to wheat revolution, developing susceptibility to all the three rusts. It is now realised that if the gains made in wheat production are to be consolidated, stabilisation of production by breeding varieties, resistant to rusts is essential. A recognised way of achieving the objective of breeding durable resistance is the incorporation of diverse genes for resistance into a variety. Rational planning of an effective breeding programme in this direction requires :

(*a*) Information on the effectiveness of known genes for resistance against rust races prevalent in India and (*b*) Identification of genes for resistance in parents to be used in the crossing programme. Since information available on both the points in the context of Indian rust flora is meagre, a programme of genetics of rust resistance is initiated at this Institute. Information on *Sr* genes, effective against Indian stem rust races is summarised in this report.

From surveys of rust infection and analysis of samples, collected from different parts of the country, race 21 (Singh, personal communication) of stem rust is found to be widely prevalent in India. For virulence, races 15, 15 C, 34 A, 42, 42 B and 122 rank important, for the varieties under cultivation in India. A logical strategy for breeding varieties that will give a rust free crop is to incorporate genes, for resistance against the 7 races listed above. Perusal of the rust reaction pattern recorded in Table I indicates that combination of genes Sr 5, Sr 6, Sr 8, Sr 9 *b* and Sr 11 will effectively meet this purpose. In fact it will provide a fair degree of overlap for resistant genes against an individual race. A combination of *Sr* 6 and *Sr* 9 *b*, for example, will provide two genes for resistance against races 11, 15 C, 21, 40 and 117 A. Rust sample analyses, during the last two years have shown that race 21 has constituted 50% of the collected samples in 1971–72 and as much as 90% in 1972–73 (Singh, personal communication). The desirable combination of genes suggested in this report (*i.e.*, *Sr* 5, 6, 8, 9 *b* and 11) will provide 5 different genes for resistance against this most prevalent stem rust race. From studies on adult plant resistance, a similar view has been expressed[1] that a combination of *Sr* 11 and other genes may impart a fair degree of field tolerance. It is also seen that

TABLE I

Infection types on monogenic Sr *lines and stocks with known gene(s) when inoculated with 16 Indian* P. graminis *races*

Line/Stock	RACE															
	11	14	15	15C	21	24	34A	40	42	42B	117	117A	122	184	194	222
Hope Mq^{10} (*Sr*1 or *Sr*9*d*) ..	0	;1	4	4	4	4	3	4	4	3	4	;1+	4	4	4	3
Thatcher Mq^{10} (*Sr*5) ..	4	0;	4	4	0;	3^{n}	4	4	0;	4	0;	;1	4	0	;1–	3
Kenya 58 Mq^{10} (*Sr*6) ..	0	4	3	;1	2, 3^{n}	4	3–4	0;1	1;	;1–	4	0;	4	4	3	3
Egypt Na 101 Mq^{11} (*Sr*7) ..	0	3	3	4	4	4	3	3	3	4	NT	3	4	4	NT	3
I *Sr7b* — Ra ..	3	;1	3	4	4	4	3	3	4	4^{c}	4	3	4	4	4	3
I *Sr*8 — Ra ..	4	3	3	3^{c}	2	3	4	3	4	3	NT	0	2, 2–	0;	NT	3
Red Egyptian Mq^{6} (*Sr*9a) ..	0	1	4	4+	4	4	4	4+	4	4	4	3	4	4	3–4	3
Kenya 117A Mq^{6} (*Sr*9b) ..	..	..	0;	;+	;–2	..	..	1	4	NT	..	;1	..	..	..	..
Gammenya (*Sr9b*) ..	0;	4	..	..	..	3–	;1–	..	..	NT	0;	..	3	0	;1	0;
Egypt Na 95 Mq^{4} (*Sr*10) ..	4	;1–	4	3	3–4	1, 3^{n}	3	4	1, 3	4^{c}	4	0;	4	4^{c}	3–4	3
Lee Mq^{10} (*Sr*11) ..	4	2–2≡	4	4	;1–	3	3	;1	2=2≡	4	4	4	4	4	0; 1	0; 1
Marquis (*Sr*12) ..	NT	NT	NT	4	4	NT	NT	4	NT	NT	NT	NT	4	NT	NT	NT
Khapstein Mq^{10} (*Sr*13) ..	2	2–2≡	3	3	4^{n}	3–4^{n}	3	3	4	4^{n}	4	;1	4	3	;1	3
Khapstein Mq^{10} (*Sr*14) ..	0	;1–	4	4	4	4^{n}	4	4	4	4^{c}	4	2=2≡	4	0	4	3
Thew (*Sr*15) ..	4	0;1	3	1·3	4	3–4	4	1, 3	3	4^{c}	0	0; 2–	4	3	3–4	3
I *Sr*16 — Ra ..	4	4	4	4	4	3	4	4	3–4	4	4	4^{n}	4	4	4	3
Spica (*Sr*17) ..	3	1^{c}	3	X	3	3	4	4	3	4^{c}	0	;1–	4	0	0	3

0 = Immune; 0; = Nearly immune; 1, 1^{c}, 1– and 1+ = Very resistant; 2 ≡ 2 =, 2–, 2 and 2+ = Moderately resistant; 3, 3^{c}, 3^{n} = Moderately susceptible; (4, 4^{c}, 4^{n} and 4– = Very susceptible; X = Mesothetic; NT = Not tested.

among the known genes tested for resistance, the genes *Sr* 1, *Sr* 7 *b*, *Sr* 9 *a*, *Sr* 13, *Sr* 14 and *Sr* 16 are largely ineffective for races of stem rust important, at the moment, in India.

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1. Rao, M. V., *Ind. J. Genet.*, 1970, 30, 59.

LEAF SPOT OF *PRUNUS CORNUTA* STEUD. INCITED BY *POLYSTIGMA RUBRUM* (PERS. EX. FR.) DC.

A SEVERE red spot on the leaves of *Prunus cornuta* Steud. was noticed at Khadrala (H.P) in June, 1974. *Prunus cornuta* Steud. a deciduous species growing at elevation of 2,000 metres to 3,033 metres has a good potentiality as root stock for cherry, as grafts on it make vigorous growth as compared to those on *Pr. cerasoides* D. Don. which is already in use as rootstock for cherry.

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Polystigma rubrum, being widely distributed pathogen in several countries, is reported to attack a large number of stone fruit plants such as Almond[1,3], Damson[15-18], Plum[1-4,7-10,12,14,19], *Prunus domestica*[17], *Pr. spinosa*[5,11] Prune, Apricot[6] and *Pr. insititia*[8]. Besides, the fungus has been reported earlier on Almond[16] from India, but there is no report on *Pr. cornuta* and hence this appears to be new host record for this country.

Thanks are due to Dr. R. N. Singh, Head, Division of Horticulture and Fruit Technology and to Dr. S. P. Raychaudhuri, Head, Division of Mycology and Plant Pathology, I.A.R.I., New Delhi-12, for providing facilities and encouragement.

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---

1. Allison, J. L., *F.A.O. Plant Prot. Bull*, 1952, 1 (1), 9.
2. Boning, K., *Landw. Jb. Bayern.*, 1952 21 Supp. 91.
3. Bremer, H., Ismen, H. and Korel, G., *et al.*, *Rev. Fac. Sci., Univ., Istanbul, Ser. B*, 1947, 13 (2), 122.
4. Chen, Y. H., Liu, F. C. and Chiang, K. C., *Zhibings, Zhishi*, 1958, 2 (2), 10.
5. Durrieu, G., *Bull. Soc. Hist. Nat.*, Toulouse, 1956, 91 (3–4), 307.
6. Esfandiari, E., *Ent. Phytopath. appl.*, Tehran, 1947, 5, 21.
7. Estifeyeff, P. G., *Phamphlet-Djetyssouy. Plant. Prot.*, Station, Almata, 1925, p. 14.
8. Fetch, T., *Trans. Brit. mycol. Soc.*, 1943, 26 (1–2), 53.
9. Khabiri, E., *Rev. Myco.*, 1952, 17 (2), 154.
10. Krasov, L. I., *Byull. glave. bot. sada*, Moskra, 1960, 87, 10.
11. Kijajic, R., *Rev. Res. Fac. Agr.*, Beograd, 2 (1), 119.
12. Lehoczky, J., *Kerteoz Szolesz, (Ann. Acad. Hort. Vitic) Bundapest.*, 1957, 21, 3.
13. Stojanovic, D. and Kostic, B., *Zasht. Bilja*, 1956, 37, 91.
14. Suta, V., *Revta. Hortic Vitic.*, 1969, 18 (1), 70.
15. Trifonova, Vera, *Phytopath. Zeitsch.*, 1934, 7 (1), 73.
16. Vasudeva, R. S., *Sci. Rep. Agri. Res. Inst.*, New Delhi, 1960, 1957–58, p. 111.
17. Wille, F., *Zentralbl. fur. Bakt. Ab.*, 1933, 2, XXXVII, 13–16, 301.
18. Yossivitch, M., *Nachrichtuber. Schadlings. Bek. Mpl.*, 1952, 7 (2), 41.
19. —, *Zasht. Bilja (Plant, Prot. Beograd)*, 1956, 37, 91.

A NEW LEAF BLIGHT OF INDIA RUBBER CAUSED BY *PHOMA GLOMERATA* (CORDA) WR. AND HOCHAPF. IN INDIA

A DISTINCTIVE leaf blight of India rubber (*Ficus elastica* Roxb.) was noted in the late winter of 1971 in New Delhi. Both the surfaces of affected leaves (young and old) bore necrotic, irregular spots, measuring several cm in diameter, and covering almost half the leaf blade (Fig. 1). The light brown to brownish-black, shrunken and thin, necrotic lesions showed in sharp contrast to the green portion of the leaf. Numerous gray-black fruiting bodies of the fungus were visible in the lesions on both the surfaces of the older leaves. Under stereobinocular microscope, a number of jet black subepidermal fruiting bodies (pycnidia) with pinkish ooze scattered all over the infected areas were clearly visible.

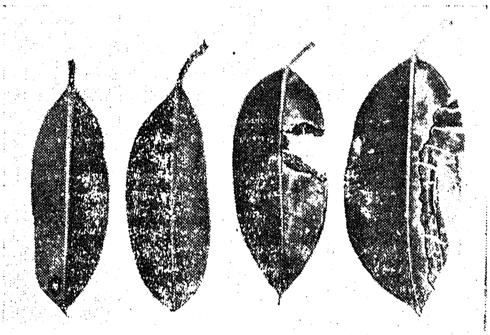

FIG. 1. Stages in the development of leaf blight disease of India rubber.

Repeated isolations of the fungus on Potato Dextrose Agar (PDA) confirmed the association of *Phoma glomerata* (Corda) Wr. and Hochapf. with the lesions. The fungus is known to cause a blight of grapevine and its blossoms and also as a secondary invader causing rot of tomato, potato, etc[1,2].

The pathogenicity of the fungus (a ten-day old culture) was tested on three year old, potted plants by spray method. First symptoms were observed 48 hours after spraying on the leaves. However, no infection was observed on petioles and stem. Reisolations from all the infected tissues on PDA repeatedly yielded *P. glomerata.*

The occurrence of *Phoma glomerata* (Corda) Wr. and Hochapf. appears to be a new report for India and India rubber as its host. The fungus culture (Accession No. 1730) has been deposited with the Type Culture Collection of the Division of Mycology and Plant Pathology, Indian Agricultural Research Institute, New Delhi.

The authors are grateful to Dr. S. P. Raychaudhuri, for his keen interest and encouragement. Thanks are also due to Dr. J. N. Kapoor, Mycologist, for his help in identification of the fungus.

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1. Commonwealth Mycological Institute : *Descriptions of Pathogenic Fungi and Bacteria,* C.M.I., Kew, Surrey, England, 1967, Paper No. 134.
2. Boerema, G. H., Dorenbosch, M. N. J. and Van Kestern, H. A., "Remarks on species of *Phoma* referred to *Peyronallae," Persoonia,* 1965, 4 (1), 47.

SILVER JUBILEE OF THE DEPARTMENT OF PHARMACY, ANDHRA UNIVERSITY, WALTAIR

The Silver Jubilee Celebrations of the Department of Pharmacy, Andhra University, will be held during the last week of April, 1976. A Seminar on Recent Advances in the Chemistry and Pharmacology of Indian Plant Drugs will be held in this connection. Further particulars may be obtained from Prof. E. Venkata Rao, Department of Pharmaceutical Sciences, Andhra University, Waltair.

among the known genes tested for resistance, the genes *Sr* 1, *Sr* 7 *b*, *Sr* 9 *a*, *Sr* 13, *Sr* 14 and *Sr* 16 are largely ineffective for races of stem rust important, at the moment, in India.

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---

1. Allison, J. L., *F.A.O. Plant Prot. Bull*, 1952, 1 (1), 9.
2. Boning, K., *Landw. Jb. Bayern.*, 1952 21 Supp. 91.
3. Bremer, H., Ismen, H. and Korel, G., *et al.*, *Rev. Fac. Sci., Univ., Istanbul, Ser. B*, 1947, 13 (2), 122.
4. Chen, Y. H., Liu, F. C. and Chiang, K. C., *Zhibings, Zhishi*, 1958, 2 (2), 10.
5. Durrieu, G., *Bull. Soc. Hist. Nat.*, Toulouse, 1956, 91 (3–4), 307.
6. Esfandiari, E., *Ent. Phytopath. appl.*, Tehran, 1947, 5, 21.
7. Estifeyeff, P. G., *Phamphlet-Djetyssouy. Plant. Prot.*, Station, Almata, 1925, p. 14.
8. Fetch, T., *Trans. Brit. mycol. Soc.*, 1943, 26 (1–2), 53.
9. Khabiri, E., *Rev. Myco.*, 1952, 17 (2), 154.
10. Krasov, L. I., *Byull. glave. bot. sada,* Moskra, 1960, 87, 10.
11. Kijajic, R., *Rev. Res. Fac. Agr.*, Beograd, 2 (1), 119.
12. Lehoczky, J., *Kerteoz Szolesz, (Ann. Acad. Hort. Vitic) Bundapest.*, 1957, 21, 3.
13. Stojanovic, D. and Kostic, B., *Zasht. Bilja*, 1956, 37, 91.
14. Suta, V., *Revta. Hortic Vitic.*, 1969, 18 (1), 70.
15. Trifonova, Vera, *Phytopath. Zeitsch.*, 1934, 7 (1), 73.
16. Vasudeva, R. S., *Sci. Rep. Agri. Res. Inst.*, New Delhi, 1960, 1957–58, p. 111.
17. Wille, F., *Zentralbl. fur. Bakt. Ab.*, 1933, 2, XXXVII, 13–16, 301.
18. Yossivitch, M., *Nachrichtuber. Schadlings, Bek. Mpl.*, 1952, 7 (2), 41.
19. —, *Zasht. Bilja (Plant, Prot. Beograd)*, 1956, 37, 91.

A NEW LEAF BLIGHT OF INDIA RUBBER CAUSED BY *PHOMA GLOMERATA* (CORDA) WR. AND HOCHAPF. IN INDIA

A DISTINCTIVE leaf blight of India rubber (*Ficus elastica* Roxb.) was noted in the late winter of 1971 in New Delhi. Both the surfaces of affected leaves (young and old) bore necrotic, irregular spots, measuring several cm in diameter, and covering almost half the leaf blade (Fig. 1). The light brown to brownish-black, shrunken and thin, necrotic lesions showed in sharp contrast to the green portion of the leaf. Numerous gray-black fruiting bodies of the fungus were visible in the lesions on both the surfaces of the older leaves. Under stereobinocular microscope, a number of jet black subepidermal fruiting bodies (pycnidia) with pinkish ooze scattered all over the infected areas were clearly visible.

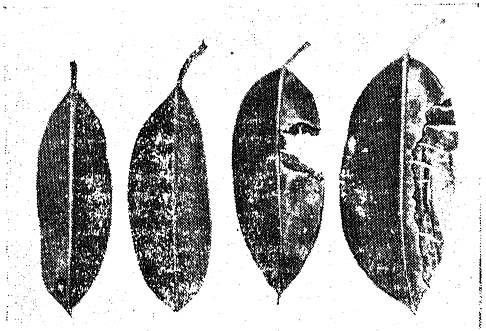

FIG. 1. Stages in the development of leaf blight disease of India rubber.

Repeated isolations of the fungus on Potato Dextrose Agar (PDA) confirmed the association of *Phoma glomerata* (Corda) Wr. and Hochapf. with the lesions. The fungus is known to cause a blight of grapevine and its blossoms and also as a secondary invader causing rot of tomato, potato, etc[1,2].

The pathogenicity of the fungus (a ten-day old culture) was tested on three year old, potted plants by spray method. First symptoms were observed 48 hours after spraying on the leaves. However, no infection was observed on petioles and stem. Reisolations from all the infected tissues on PDA repeatedly yielded *P. glomerata*.

The occurrence of *Phoma glomerata* (Corda) Wr. and Hochapf. appears to be a new report for India and India rubber as its host. The fungus culture (Accession No. 1730) has been deposited with the Type Culture Collection of the Division of Mycology and Plant Pathology, Indian Agricultural Research Institute, New Delhi.

The authors are grateful to Dr. S. P. Raychaudhuri, for his keen interest and encouragement. Thanks are also due to Dr. J. N. Kapoor, Mycologist, for his help in identification of the fungus.

Division of Mycology and
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Indian Agricultural Research
Institute, New Delhi 110 012,
September 18, 1975.

P. M. MUKEWAR.
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1. Commonwealth Mycological Institute : *Descriptions of Pathogenic Fungi and Bacteria*, C.M.I., Kew, Surrey, England, 1967, Paper No. 134.
2. Boerema, G. H., Dorenbosch, M. N. J. and Van Kestern, H. A., "Remarks on species of *Phoma* referred to *Peyronallae*," *Persoonia*, 1965, 4 (1), 47.

SILVER JUBILEE OF THE DEPARTMENT OF PHARMACY, ANDHRA UNIVERSITY, WALTAIR

The Silver Jubilee Celebrations of the Department of Pharmacy, Andhra University, will be held during the last week of April, 1976. A Seminar on Recent Advances in the Chemistry and Pharmacology of Indian Plant Drugs will be held in this connection. Further particulars may be obtained from Prof. E. Venkata Rao, Department of Pharmaceutical Sciences, Andhra University, Waltair.

SHORT SCIENTIFIC NOTES

A First Report of *Curvularia brachyspora* Boedijn Inciting Leaf-Spot Disease of Rose

During a survey for diseases caused by genus *Curvularia* several rose plants growing in the garden of Horticulture Department, Agriculture College, Poona, were found to be severely affected by leaf-spot disease. A critical microscopic observation revealed that it was due to *Curvularia* sp. The symptoms were on leaves only. There were irregular light black spots from margin to midrib. The spots increased in size, affected leaflets turned yellow, then started withering and in some cases finally shed off after drying. Isolations from such affected leaf-spots yielded a species of *Curvularia.* In artificial inoculation tests, the fungus was found to be pathogenic. Host range studies showed that the fungus was pathogenic on Bajra, Sorghum cultivars, CSH–1, CSH–2 and Swarna, Paddy, Sugarcane, blue panic (*Panicum antidotale* Retz.) Marverl 93 (*Dichanthium caricosum* A. Genus.), thin napier (*Pennisetum-polystachyon* Schult.), Grape, Soybean, Cotton (*Gossypium abroreum* L.), Ginger, Gladiolus, Pinus (*Pinus pumula* Regel), Screw pine and *Cynodon dactylon* Pers., out of thirty three hosts tested besides its own host.

The description and measurements of the test fungus very nearly resemble those of *Curvularia brachyspora* given by Ellis[1] whose measurements of conidia were 19–26 (22) $\mu \times$ 10–14 (12) μ and hence the fungus was identified as *Curvularia brachyspora* Boedijn. The leaf-spot disease of rose incited by *Curvularia brachyspora* Boedijn has been reported for the first time in India.

Plant Pathology Department, S. S. KORE.
Marathwada Agricultural University, V. P. BHIDE
Parbhani, *July* 28, 1975.

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1. Ellis, M. B., "Dematiaceous Hyphomycetes VII. *Curvularia, Brachysporium,* etc.," *Commonwealth Mycol. Inst., Mycol. Paper,* 1966, 106, 57.

Rare Occurrence of *Amphiprion percula* (Lacépède) Eggs in the Gut Contents of *Ambassis commersoni* (Cuvier)

Observations on the food and feeding habits of *Ambassis* spp. are very limited[1,2]. During our study on the feeding habits of *A. commersoni,* the stomach contents proved that this species is a plankton feeder, feeding mainly on mysids of prawn, adults of *Acetes indicus,* copepods and rarely on diatoms. However, in 2 specimens out of 15 captured (45·0–57·0 mm Std. length) during January 1975, the stomachs were filled with 200 and 290 eggs of *Amphiprion percula.* These eggs were identified by their elongated shape with an average length of 2·2 mm, a narrow perivitelline space and one big and a number of smaller oil globules[3]. Partly spawned *A. percula* were also obtained in large numbers in cast-net catches in the Vellar river mouth coinciding with the occurrence of eggs.

A. percula is a pelagic fish with demersal eggs and *A. commersoni* is a plankton feeder and it was therefore puzzling to find the eggs of *A. percula* in the stomach of *A. commersoni.* A Similar instance of *A. gymnocephalus* feeding on mullet eggs has been reported from Chilka lake[1]. May be that *A. commersoni* descended to the bottom for feeding as instances of a pelagic fish feeding on bottom organisms are not rare[4]. Problems of distinguishing between generalized and selective feeding are considerable[5]. It is difficult to decide in this instance between selective and indiscriminate feeding.

We are thankful to Dr. R. Natarajan, Director, Centre of Advanced Study in Marine Biology, for encouragement. One of us (K.V.) grateful to the U.G.C., for the award of a Junior Research Fellowship.

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Biology, JOSEPH XAVIER RODRIGO.
Annamalai University,
Porto Novo 608 502,
Tamil Nadu, India, *August* 18, 1975.

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1. Natarajan, A. V. and Patnaik, S., *Jour. mar. biol. Ass. India,* 1957, 9, 192.
2. Venkataramanujam, K., *Recent Research in Estuarine Biology,* Ed., Natarajan, R., Hindustan Publishing Corporation (I), Delhi, 1972, p. 296.
3. Delsman, H. C., *Fish eggs and larvae from the Java Sea.* Reprint, Linnaeus Press, Amsterdam, 1972.
4. Prasad, M. A. S., *Jour. mar. biol. Ass. India,* 1957, 9, 441.
5. Graham, M., *Sea fisheries,* London, 1956.

Flower Bud Rot of Marigold (*Tagetes erecta* L.) Caused by *Alternaria dianthi* Stevens and Hall in West Bangal

There are a few *Alternaria* species occurring on various parts of the marigold plant (*Tagetes erecta* L.). The two reported *Alternaria* species pathogenic to marigold, in India are *A. Zinnae* Pape[1] and *A. tagetica* Shome and Mustafee[4]. Both the species cause blighting of leaves and flowers. The infections of *A. tagetica,* have further been reported to be systemic[4]. Recently, a dry rot of developing flower buds of marigold (var. Sutton's Climax, F_1, hybrid mix.) has been seen to be caused by *Alternaria dianthi* Stevens and Hall.

Infections were obtained during the months January to March, 1975. Young, flower buds showed dry rotting with characteristic brown, scorched, necroting discolouration of the sepals and peduncle. The ray and disc florets were also infected and turned brown. At later stages the buds were shrivelled, turned deep brown to black and dried up. Symptoms were less prominent on more matured buds, which however, failed to open, and there was hardly any infection on the flowers already in bloom. A few deep brown, necrotic spots developed on the older leaves towards their margin and tips and were confluent. The symptoms were, however, more prominent on the floral parts. The fungus brought into pure culture from single spores proved to be pathogenic to the leaves and buds of marigold, on spray inoculation by conidia.

Colonies of the fungus on PDA were dark olive-brown with moderate development of air mycelium of paler colour. The conidiophores were straight to curved, septate, pale brown, upto 112 μ long and 5–8 μ broad. Conidia, formed in short chains of 2–4, were straight to slightly curved, conical to obclavate or elliptic, olivaceous brown, darker when old, measuring $13 \cdot 5$–$64\ \mu \times 8 \cdot 5$–$22\ \mu$, with a maximum of 10 transverse and 6 longitudinal septa ; Beak not very prominent, short, often swollen at the tip, concolourous with the main body of conidium.

On the basis of its salient characteristics as compared to the type descriptions of the species[2,3], the fungus was identified as *Alternaria dianthi* Stevens and Hall. The species is widely different in its conidial characters including size measurements from the other two species of *Alternaria* reported from India as pathogenic to marigold. *Alternaria dianthi* though generally is restricted to the members of caryophyllaceae, can also infect other hosts[2]. This is the first record of its occurrence on marigold (*Tagetes erecta* L.) in India. The culture is deposited with the Indian Type culture collection at IARI, New Delhi, under ac. No. 1982.

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West Bengal, *August* 18, 1975.

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1. Edward, J. C., "Leaf and infloresence blight of marigold caused by *Alternaria Zinnae* Pape," *Sci. and Cult.*, 1957, 22, 683.
2. Ellis, M. B., *Dematiaceous Hyphomycetes*, C–M–I, Kew, England, 1971, p. 608.
3. Neergaard, P., *Danish Species of* Alternaria *and* Stemphylium, Oxford Univ. Press, London, 1945, p. 560.
4. Shome, S. K. and Mustafee, T. P., "*Alternaria tagetica*, sp. nov. causing blight of marigold (*Tagetis* sp.)," *Curr. Sci.*, 1966, **14**, 370.

Additional Hosts for *Pseudomonas solanacearum* Smith

Pseudomonas solanacearum, the incitant of bacterial wilt disease, attacks an extremely wide range of host plants belonging to different families[1-3]. During the course of a study for natural occurrence of this disease on different hosts, three weed plants, *viz.*, *Phyllanthus niruri* L. (Euphorbiaceae), *Cleome monophylla* L. (Capparidaceae), and *Acanthosperma hispidum* L. (Acanthaceae) were noticed showing wilt symptoms in wilt sick plots of tomato and brinjal at Hessaraghatta. Examination of the root and stem cuttings of the infected plants revealed typical bacterial streaming from the cut ends. The pathogen was isolated on Triphenyl tetrazolium chloride agar medium[2] and the pathogenicity was confirmed on the respective host plants by stem inoculation[5] with 48 hour old culture of the isolates. These were also pathogenic on tomato (var. Pusa Ruby) and brinjal (var. Pusa Purple Long). The disease symptoms appeared late (20–25 days) in all the three weed hosts compared to tomato and brinjal (7–10 days). Under natural conditions, of infection, wilt incidence was more on *P. niruri* and *C. monophylla* than on *A. hispidum*. Further, *P. niruri* was also found as a symptomless carrier of the pathogen.

Out of the three weed hosts, *P. niruri* has been reported earlier as a host under natural conditions[4] and our findings are in conformity with this report. The other two weeds, *C. monophylla* and *A. hispidum* are new additions to the host range of *Pseudomonas solanacearum.*

The authors are grateful to Dr. G. S. Randhawa, Director, for his interest and encouragement. Thanks are also due to Dr. D. Leela of Plant

Physiology Division, for help in the identification of the weeds.

Indian Institute of
Horticultural Research,
Bangalore, *August* 21, 1975.

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1. Kelman, A., *North Carolina Agr. Exp. Sta. Tech. Bull.*, 1953, 99, 48.
2. —, *Phytopathology*, 1954, 44, 693.
3. Sulladmath, V. V., Hedge, R. K., Patil Kulkarni, B. G. and Hiremath, P. C., *Curr. Sci.*, 1975, 44, 286.
4. Vander Wolk, P. C., *Indische Mercurr.*, 1914, 37, 647.
5. Winstead, N. N. and Kelman, A., *Phytopathology*, 195, 42, 628.

Occurrence of Bajara Mosaic in U.P. (India), with *Myzus persicae* Sulz. as an Additional Vector

In July, 1973, a mosaic disease (incidence of 3–6%) has been observed on Bajara [*Pennisetum typhoides* (Burm. f.) Stapf. and C. E. Hubb] near Faizabad (U.P., India). The infected plants were stunted, with the leaves showing severe mosaic consisting of broken, chlorotic streaks of varying sizes.

The disease was sap transmissible to *Cynodon dactylon* L., *Dactyloctenium aegyptiacum* Willd., *Eleusine coracana* L., *Panicum miliaceum* L., *Paspalum scrobiculatum* L., *Setaria italica* (L.) Beauv., *Oryza sativa* L. var. IR–8, and *Zea mays* L. But attempts to infect *Arundo donax* L., *Triticum aestivum* L., *Hordeum vulgare* L., *Saccharum officinarum* L. var. B.O. 14, CO. 740, *Nicotiana tabacum* L., and *Phaseolus vulgaris* L. were unsuccessful.

The virus showed dilution end point in between 1 : 500–1 : 1000, and a thermal inactivation point in between 55–60° C (when heated for 10 minutes). Longevity '*in vitro*' varied from 12–16 hours at room temperature (30 ± 1° C).

The virus was transmitted in a stylet-borne manner from artificially infected maize to maize by the aphid *Rhopalosiphum maidis* Fitch. up to 40% and by *Myzus persicae* Sulz. up to 30%. However, *Aphis craccivora* Koch. could not transmit this virus.

On the basis of symptomatology, host range, physical properties and aphid vectors, the virus has been identified as Bajara Mosaic Virus reported by Seth *et al.*[1]. *Myzus persicae* Sulz. has now been shown to be an additional vector.

The author is thankful to Dr. R. S. Tiwari, Principal, K. S. Saket, Post-Graduate College, Faizabad (U.P.), for providing the necessary facilities during the present investigation.

Department of Botany,
K.S. Saket Post-Graduate College,
Faizabad (U.P.), *August* 27, 1975.

CHAUDHARY A. K. SINGH.

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1. Seth, M. L., Ray Chaudhuri, S. P. and Singh, D. V., *Indian J. Agric. Sci.*, 1972, 42, 322.

A New Post-Harvest Disease of Pomegranate in India

During the survey of different fruit markets of Rajasthan, large number of pomegranate fruits were found to be rotting due to *Alternaria solani* in November, 1974. It was observed that this fungus causes considerable damage during storage and transit. Generally the infection was common in cracked fruits. The symptom of the fruit rot was characterised by the development of light-brown spots, which gradually changed to dull-brown. Later on, due to severe infection, the fruits became pulpy. The grains of the infected fruits became greenish and eventually turned dark due to sporulation of the fungus. Rotten fruits emitted fermented odour.

Pathogenicity of the organism was confirmed by inoculating the fruits by Granger and Horne's method[1] and also by spraying the conidial suspension of the organism over the injured and uninjured fruits. Only injured fruits developed typical symptoms. Reisolations made from artificially infected fruits yielded pure cultures of *Alternaria solani.* This fungus disease of pomegranate has not been reported in literature so far.

The senior author is grateful to C.S.I.R., New Delhi, for the award of Post Doctoral Fellowship and to Prof. H. C. Arya, Head, Department of Botany, for providing laboratory facilities.

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1. Granger, K. and Horne, A. S., *Ann. Botany*, 1924, 38, 212.

REVIEWS AND NOTICES OF BOOKS

Green's Functions for Solid State Physicists. By S. Doniach and E. H. Sondheiner. (W. A. Benjamin, Inc., Reading, Mass.), 1974. Pp. xix + 266. Price $ 19.00.

For nearly fifteen years by now, Messrs. Benjamin, Inc., have been bringing out a number of lecture notes and reprints in a series entitled 'Frontiers in Physics' under the General Editorship of David Pines. The books are intended to be topical and expected to summarize the present status of rapidly developing fields and make the lecture notes available to a wider audience and the volumes have been, in general, found useful by the physics community. The present book is the 44th, based on a lecture course by S. Doniach.

This book, authors hope, 'will appeal to beginning graduate students in theoretical solid state physics as a first introduction to more comprehensive or more specialized texts and also to experimentalists who would like a quick, if impressionistic view of the subject. A basic knowledge of solid state physics and quantum mechanics at undergraduate honours or first year graduate level is assumed'.

The hope is not fully realized. As an introduction to the subject for self-study, for initiated, the book appears to be rather heavy reading. The book would be useful as a supplement to lectures, though not qualifying as a text book. Several topics of interest are dealt with and the power of the Green's function method for tackling many body problem is brought out. The link with experiment is very trenuous and limited to a few graphs here and there which do not add much to the understanding of the subject matter. Perhaps the book has lost the informal character of a lecture notes but not replaced by other aids to get a quick over-view of the subject, its use and limitations. It would be very useful if such books have a suitable introduction, not necessarily by the authors, but perhaps by the general editor, who can in a short account, preferably by pictorial, tabular or tree form, bring out the place of the topic in the overall scheme of things, thereby adding perspective. In a lecture this is easily supplied by the lecturer through questions and discussions but generally lost in cold print.

The book has a useful bibliography and a readable historical note on George Green though one wonders if George Green were to come alive, he would find the contents of this book understandable without further aid! G. SURYAN.

Petroleum and Global Tectonics. Edited by Alfred G. Fischer and Sheldon Judson. (Princeton University Press), 1975. Pp. 322. $ 16.50.

This volume contains a collection of nine papers presented by eminent geologists, geophysicists and geochemists at a conference held in honour of Prof. Hollis Hedberg at Princeton University. The role of tectonic features in the generation and accumulation of petroleum has long been known, but these ideas were derived mostly from the general tectonic studies of the earth's continental masses. In recent years, the geological and geophysical studies of the sea floor, which constitutes two-thirds of the total global area, have radically altered our ideas of the tectonic features of the entire earth. It is therefore both timely and appropriate that the generation, accumulation and distribution of petroleum in relation to global tectonics (in particular Plate Tectonics) were considered and reviewed by several authorities in a conference, and the proceedings made available so that the large body of earth scientists could share this knowledge.

The first paper on "Over-view of Plate Tectonics" by Edward C. Bullard furnishes a masterly, succinct account of the present status of knowledge concerning plate tectonics, and points out generally the relation between petroleum and plate tectonics.

There are two papers, in a way supplementing each other, on the varied aspects of sedimentary basins. In his paper on "the Origin and Growth of Basins" Alfred G. Fisher stresses the different mechanisms that explain the development of various types of sedimentary basins and concludes that plate tectonics and other geological insights provide a variety of plausible mechanisms which trigger each other and interact, so that no basin can be regarded as due to any one cause alone. Joseph R. Curray dealing with "Marine sediments, Geosynclines and Orogeny" offers a classification of geosynclines based on the present sedimentation patterns related to plate tectonics. Of particular interest to petroleum geologists in India is a diagrammatic depiction, he has presented regarding the scraping of sediments from the Bengal Fan on the subducting Indo-Australian plate, and the anticlinal structure of the Andaman Islands.

Two papers, one by David Kinsman entitled "Rift Valley Basins and Sedimentary History of Trailing Continental Margins", and the other by James D. Lowell and his collaborators on "Petroleum and Plate Tectonics of the Red Sea", demonstrate the close relationship between plate tectonics and the control on the occurrence of petroleum.

Two papers deal with geothermal aspects : W. Jason Morgan on "Heat Flow and Vertical Movements of the Crust" and H. Douglas Klemme on "Geothermal Gradients, Heat Flow and Hydrocarbon Recovery" deal comprehensively with the dissipation of the heat generated in the interior of the earth, and the geological work that the heat transfer accomplishes. It has been pointed out that the basins with relatively high heat flow—such as the mobile continental margins—offer optimal conditions for the generation, migration and accumulation of petroleum.

James Gordon Erdman in his paper on "Geochemical Formation of Petroleum" reviews the general processes by which organic fractions of the seediments are transformed by inorganic processes into hydrocarbons. He points out that in intercontinental areas lie the best remaining prospects for discovery of petroleum deposits.

Finally, J. D. Moor in his paper on "Distribution and Geological Character of Giant Oil Fields" presents the results of a very interesting statistical study of the data on 198 giant oil fields (*i.e.*, fields expected to yield more than 500 million barrels of oil) which constitute a little over two-thirds of the world's known reserves. His findings are (1) that more giant fields were found in the last decade than in any of the previous ten-year periods, (2) the Tertiary-Mesozoic formations have yielded the most prolific producers, and (3) anticlinal structures furnish the most efficient types of trapping mechanism. His prognostication is that the potential oil yet to be found in the world may be estimated at 800–900 billion barrels. Assuming that 75% of this quantity will be found in giant fields, he surmises some 200–300 giants remain yet to be found in the world—a welcome hope indeed which will cheer up all exploration geologists and geophysicists and the oil industry men.

Each paper is amply illustrated and the book as a whole is a valuable treatise adding to the literature not only on petroleum geology but also on plate tectonics. It is remarkable that the revolutionary ideas regarding plate tectonics, based on large scale studies of the ocean floor, find support from the large volume of details gathered during petroleum exploration, and that the plate tectonics theory in turn has influenced current thinking on further exploration. The Editors have done a good job in compiling the papers and giving a short introductory para on each of them to help the readers in following these valuable contributions. The printing and get-up of the book is excellent.

M. B. R. Rao.

Practical Plant Pathology (Revised Edition). By M. N. Kamat. (Prakash Publishing House, Poona-2), 1971. Pp. 1–117. Price Rs. 13.

This is a second edition after the publication of the first edition in 1953 on practical Plant Pathology. After brief prefaces to the two editions and short life sketches of such great Pathologists as Anton deBary, Juleus Kuhn, Robert Koch and Louis Pasteur, they are followed by a series of disease causing Fungi from Phycomycetes, Ascomycetes, Basidiomycetes, Fungi Imperfecti and Schizomycetes. The several plant diseases affecting the well-known crops of India are described in a detailed manner with suitable line drawings or photographs and these are in the form of Exercises. The different aspects of Plant Pathology are dealt with and in almost every case information is provided on symptamology, etiology, life cycle and control. Suitable references are cited under each exercise for additional information. The entire approach has been to serve as a practical guide to students who study plant pathology. A book of this nature will be particularly useful in agricultural universities and others interested in plant pathology.

K. Subramanyam.

ANNOUNCEMENTS

Indian Notional Science Academy, New Delhi

At the Annual General Meeting of the Academy held on Friday, the 10th October 1975, Dr. B. P. Pal, FRS, was re-elected as the President. The other officers re-elected for the ensuing year were : Vice-Presidents : Dr. K. Venkataraman, Natioal Chemical Laboratory, Poona, and Prof. M. R. N. Prasad, Head, Department of Zoology, University of Delhi ; Treasurer : Dr. A. R. Verma, Director, National Physical Laboratory, New Delhi ; Foreign Secretary : Dr. B. D. Nagchaudhuri. Vice-Chancellor, Jawaharlal Nehru University, New Delhi ; Secretaries : Dr. A. B. Joshi, Director, Indian Agricultural Research Institute, New Delhi and Prof. A. N. Mitra, Professor of Physics, University of Delhi; Editors of Publications : Professor L. S. Kothari, Professor and Head of the Department of Physics, University of Delhi, and Prof. K. N. Saxena, Professor of Zoology, University of Delhi, Delhi.

The other members of Council elected were: Prof. B. K. Bachhawat, Christian Medical College, Vellore; Dr. V. G. Bhide, National Physical Laboratory, New Delhi; Prof. A. Bose and Prof. P. C. Datta, from Indian Association for the Cultivation of Science, Calcutta; Prof. D. Lal, Director, Physical Research Laboratory, Ahmedabad; Dr. Nityanand, Director, Central Drug Research Institute, Lucknow; Dr. N. Parthasarathy, 16, Satyanarayana Avenue, Boat Club Road, Madras; Dr. K. G. Ramanathan, Tata Institute of Fundamental Research, Bombay; Dr. R. Ramanna, Director, Bhabha Atomic Research Centre, Bombay; Dr. S. Ramaseshan, National Aeronautical Laboratory, Bangalore, Dr. C. S. Ramaswamy, Director, Structural Engineering Research Centre, Madras; Prof. C. N. R. Rao, Dean of Research and Development, Indian Institute of Technology, Kanpur; Prof. Ajit Kumar Saha, Presidency College, Calcutta; Prof. A. K. Sharma, University of Calcutta, Calcutta; Dr. S. N. Sarkar, Indian School of Mines, Dhanbad; Dr. A. Sreenivasan, Tata Memorial Centre, Bombay; Dr. K. Sundaram, Director, Biomedical Group, Bhabha Atomic Research Centre, Bombay; Dr. M. V. K. Udupa, Director, Central Electrochemical Research Institute, Karaikudi; Dr. D. S. Kothari, Immediate Past President, A-3 Chatra Marg, Delhi; Prof. R. N. Tandon, President, National Academy of Sciences, India, Allahabad; Dr. N. N. Das Gupta, 44, Hazara Road, Calcutta; Dr. A Ramachandran, Secretary to the Government of India, Department of Science and Technology, New Delhi.

Fellows Elected:

Prof. U. Aswathanarayana, University of Saugor, Sagar, M.P., Prof. K. S. Bhargava, Gorakhpur University, Gorakhpur; Prof. P. K. Bhattacharyya, Indian Institute of Science, Bangalore; Dr. P. K. Iyengar, Bhabha Atomic Research Centre, Bombay; Dr. S. S. Joshi, CIBA Research Centre, Bombay; Prof. G. K. Manna, Kalyani University, Kalyani; Dr. N. K. Notani, Bhabha Atomic Research Centre, Bombay; Dr. I. C. Pande, Punjab University, Chandigarh; Prof. K. R. Parthasarathy, University of Bombay, Bombay; Prof. G. S. Sanyal, Indian Institute of Technology, Kharagpur; Prof. M. V. C. Sastri, Indian Institute of Technology, Madras; Prof. M. M. Sharma, University of Bombay, Bombay; Prof. B. V. Sreekantan, Tata Institute of Fundamental Research, Bombay; Dr. S. Sriramachari, Safdarjang Hospital, New Delhi; Prof. S. K. Trehan, Punjab University, Chandigarh.

Foreign Fellows Elected:

Prof. John Bardeen (USA), Prof. Alfven Hannes Glof Gosta (Sweden), Prof. Robert Hanbury Brown (Australia), Dr. Jacques Lucien (France), Prof. Rudolf Mössbauer (France), Prof. Ralph Riley (Cambridge).

'Science Academy Medals for Young Scientists' were awarded to the following:

Dr. S. K. Kataria, BARC, Bombay; Dr. J. Lakshmana Rao, S. V. University, Tirupati; Dr. K. S. Dhindsa, Director, Haryana Agricultural University, Hissar; Dr. M. L. Munjal, Indian Institute of Science, Bangalore; Dr. Shamsuddin, B.H.U., Varanasi; S. Krishnaswami, Physical Research Laboratory, Ahmedabad; Dr. Naresh Kochhar, Chandigarh; Dr. N. C. Bhattacharya, Panjab University, Chandigarh; Dr. A. S. Khalatkar, Nagpur University, Nagpur, Dr. S. C. Lakhotia, Ahmedabad, Dr. A. A. Nagroo, Maulana Azad Medical College, New Delhi; Dr. Manju, Ray, Jadavpur University; Calcutta.

Prof. K. S. Krishnan Memorial Lectureship (1975) was awarded to Dr. A. P. Mitra and Prof. Biresh Chandra Guha Memorial Lectureship to Prof. Jagannath Ganguly.

Award of Research Degrees

University of Saugar, Sagar, has awarded the Ph.D. degree in Zoology to Shri Devendra Nath Saksena.

Mahatma Phule Krishi Vidyapeeth has awarded the D.Sc. degree to Prof. M. N. Kamat, President, Mycological Society of India and Head, Department of Mycology and Plant Pathology, Maharashtra Association for the Cultivation of Science, Poona-4.

Berhampur University, Berhampur, has awarded the Ph.D. degree in Chemistry to Shri Prakash Chandra Patnaik; and Ph.D. degree in Mathematics to Shri Tarini Charan Pand.

Karnatak University, Dharwar, has awarded the Ph.D. degree in Mathematics to Shri Vasudev Shridhar Shetiya, Shri S. B. Chikkodimath, Shri Vithal Narayanrao, Bhave.

Karnatak University, Dharwar, has awarded the Ph.D. degree in Physics to Shri G. C. Chikkur.

Karnatak University, Dharwar, has awarded the Ph.D. degree in Chemistry to Shri Raghavendra Jeevanrao Deshpande, Shri V. V. Kanabur.

Karnatak University, Dharwar, has awarded the Ph.D. degree in Agriculture to Shri H. S. M. Channabasaiah, Shri Tippannavar Mallappa Basappa.

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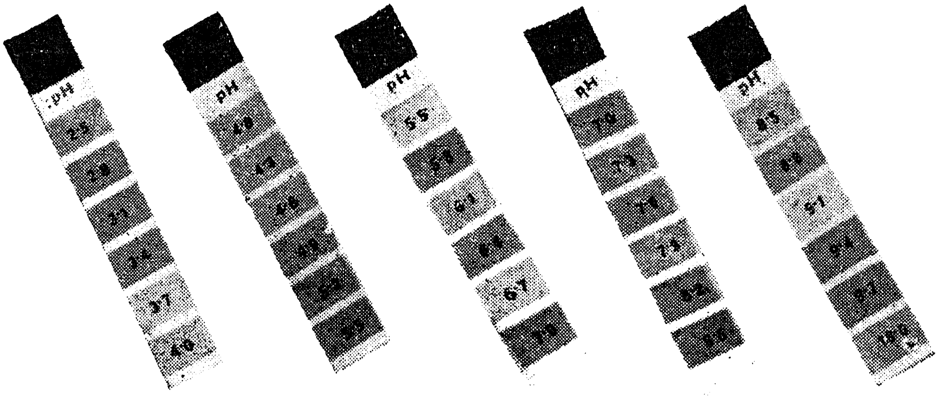

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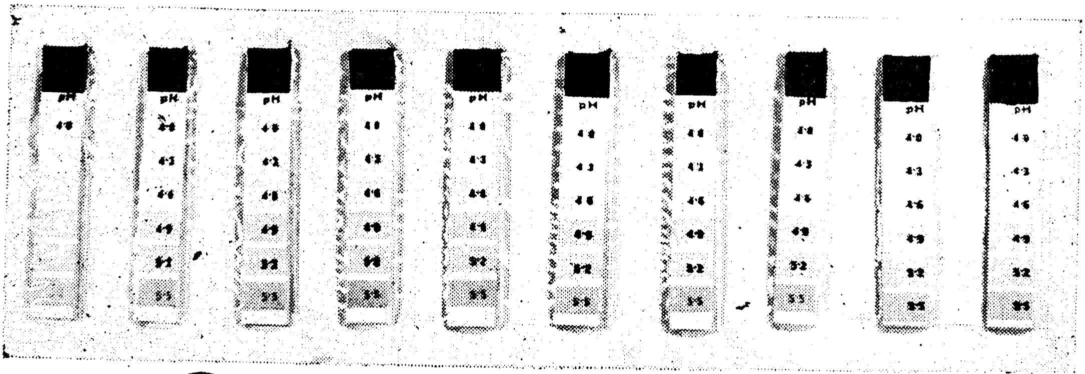

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COMPLEXES OF Cu(I) AND Zn(II) WITH SOME N-ARYL, N′-2-(5-HALO-PYRIDYL) THIOUREAS

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METAL complexes of the substituted thioureas having a heterocyclic group as one of the substituents have been studied by many workers[1-4]. In these complexes pyridyl nitrogen and thiocarbonyl sulfur act as donor atoms, giving rise to a stable six membered chelate ring. The present paper reports the studies made on the complexes of Cu (I) and Zn (II) with some N-aryl, N′-2-(5-halo-pyridyl) thioureas.

The following are the substituted thioureas that were chosen :

(*i*) N-phenyl, N′-2-(5-chloro-pyridyl) thiourea (PCT),
(*ii*) N-phenyl, N′-2-(5-bromo-pyridyl) thiourea (PBT),
(*iii*) N-phenyl, N′-2-(5-iodo-pyridyl) thiourea (PIT),
(*iv*) N-*o*-tolyl, N′-2-(5-chloro-pyridyl) thiourea (TCT),
(*v*) N-*o*-tolyl, N′-2-(5-bromo-pyridyl) thiourea (TBT), and
(*vi*) N-*o*-tolyl, N′-2-(5-iodo-pyridyl) thiourea (TIT).

EXPERIMENTAL

Preparation of the Cu (I) Complexes.—0·01 mole of $CuCl_2 \cdot 2H_2O$ (BDH, AnalaR) in ethanol was added slowly and with stirring to a hot solution (0·025 mole) of the substituted thiourea in *n*-butanol. The complex was precipitated just after the addition. It was filtered, washed with butanol and dried in air oven at 60–70° C.

A hot solution (0·01 mole) of $Zn(ClO_4)_2 \cdot 6H_2O$ in *n*-butanol was added with stirring to a boiling solution (0·025 mole) of the ligand in the same solvent. The mixture was concentrated and cooled. The precipitate was filtered, washed with *n*-butanol and dried at 60–70° C.

The composition of the complexes was determined by the estimation of the elements, N, S, Cl [ClO_4 in case of Zn (II) complexes] and the metal. Copper and zinc were estimated by the pyridine method[6], as $[M(C_5H_5N)_4](CNS)_2$. The other elements were estimated by standard methods.

The conductance measurements were made on a conductivity meter, type LBR, WTW, Germany, using a dip type cell. Ultra-violet spectra were obtained on a UV–VIS Perkin Elmer Hitachi Spectrophotometer (Model–139) using 0·5 cm matched quartz cuvettes. Infrared spectra, in nujol-mull were recorded on a Perkin Elmer Grating Infrared Spectrophotometer (Model 237–B).

RESULTS AND DISCUSSION

The Cu (I) complexes, white in colour, were soluble in chloroform, dimethylformamide, nitrobenzene, pyridine and dioxane. These complexes, on the basis of elemental analysis can be represented by the general formula $CuClL_2$, where L is the ligand. The Zn (II) complexes were also white in colour and were soluble in acetone, pyridine, tetrahydrofuran and dimethylformamide. The elemental analysis of these compounds revealed their composition to be $Zn(ClO_4)_2L_2$. The results of elemental analysis along with their melting points, molar conductances and magnetic moments have been recorded in Table I.

The Cu (I) Complexes.—The composition of these complexes shows that Cu is present in + 1 oxidation state. The reduction of Cu (II) to Cu (I) is a peculiar property of thioureas and has been observed by earlier workers also[1-3].

The low molar conductance range (0·04–0·22 mhos, *cf.* Table I) shows that these complexes are non-electrolytes, having chlorine coordinated to the metal ion.

Two bands are exhibited in the uv spectra of these complexes which have been assigned to thiocarbonyl $\pi - \pi^*$ transition (~ 275 nm) and pyridyl $\pi - \pi^*$ transition (~ 310 nm)[4]. On comparison of these spectra with those of the ligands it has been found that the λ_{max} of thiocarbonyl $\pi - \pi^*$ transitions is shifted upto 10 nm to shorter wavelengths. In the case of ligands E_{max} for thiocarbonyl $\pi - \pi^*$ transition is found to be greater than that for pyridyl $\pi - \pi^*$ transition but in the case of Cu (I) complexes the order is reversed (*cf.* Table II). These observations are indicative of strong interactions between the ligand and the metal ion. Cu (I) is a soft acid according to the Pearson's classification[7] and belongs to the (*b*) class of metals according to Ahrland *et al.*[8]. Therefore its coordination with a doft donor, like thiocarbonyl sulfur with strong π–interactions is quite obvious. This however, has not been reported in the case of complexes of substituted thioureas by earlier workers[1-3].

TABLE I

Copper (I) chloride and Zinc (II) perchlorate complexes of some N-aryl, N′-2-(5-halo-pyridyl) thioureas

Complex	m.pt. (°C)	Molar conductance (mhos)	Magnetic moment (BM)	% Metal		% Nitrogen		% Sulfur		% chlorine perchlorate	
				Calculated	Found	Calculated	Found	Calculated	Found	Calculated	Found
$CuCl(PCT)_2$	246d	0·10	Diamagnetic	12·77	12·65	13·42	13·32	12·87	12·75	17·01	16·90
$Zn(PCT)_2(ClO_4)_2$	208d	70·2	do.	8·26	8·15	10·61	10·41	8·09	8·01	25·15	25·02
$CuCl(PBT)_2$	248d	0·04	do.	8·88	8·75	11·75	11·62	8·95	8·82	4·97	4·85
$Zn(PBT)_2(ClO_4)_2$	210d	72·9	do.	7·43	7·31	9·54	9·38	7·27	7·12	22·60	22·42
$CuCl(PIT)_2$	239d	0·18	do.	7·85	7·70	10·39	10·18	7·91	7·80	4·39	4·12
$Zn(PIT)_2(ClO_4)_2$	221d	74·0	do.	6·71	6·51	8·62	8·51	6·57	6·47	20·42	20·30
$CuCl(TCT)_2$	230d	0·10	do.	9·71	9·61	12·84	12·64	9·79	9·60	16·29	16·09
$Zn(TCT)_2(ClO_4)_2$	215d	73·1	do.	7·98	7·85	10·75	10·05	7·81	7·65	24·29	24·02
$CuCl(TBT)_2$	224d	0·08	do.	8·55	8·43	11·31	11·11	8·61	8·41	4·78	4·65
$Zn(TBT)_2(ClO_4)_2$	210d	76·2	do.	7·20	7·02	9·25	9·15	7·05	6·91	21·91	21·75
$CuCl(TIT)_2$	222d	0·22	do.	7·59	7·45	10·03	9·95	7·65	7·48	4·24	4·15
$Zn(TIT)_2(ClO_4)_2$	210d	79·0	do.	6·33	6·21	8·38	8·29	6·39	6·32	19·86	19·73

d = decomposes.

In the spectra of the complexes (Table III), the ν (N—C—N+C=S) and ν (C=S) bands occurring at ~ 1060 cm^{-1} and ~ 720 cm^{-1} respectively[4] are either shifted to lower frequencies or are reduced in their intensities. The band assigned to ν (C=C+C=N) mode is shifted in an irregular fashion which precludes any unambiguous assertion of metal ligand bonding through the pyridyl nitrogen, which however is suggested by the uv spectra.

On the basis of these observations the complex can be considered five coordinated (2 pyridyl nitrogens, 2 thiocarbonyl sulphurs and 1 chlorine.

The Zn (II) Complexes.—The molar conductances of these complexes fall in the range 70–80 mhos predicting these to be 1 : 1 electrolytes. But the ir spectra of these complexes do not show the characteristic bands of coordinated perchlorate[9]. Thus, the complexes should be 1 : 2 electrolytes. It has been observed by Dutta Ahmed and Mandal[1] that in non-aqueous solvents the conductances of 1 : 2 electrolytes are less than the normal values.

The uv spectra of these complexes (Table II) show little variation from the corresponding ligand spectrum.

The band at ~ 1600 nm assigned for ν (C=C+C=N) mode is found to be shifted to lower frequencies showing thereby bonding through pyridyl nitrogen. The ν (N—C—N+C=S) and is either missing, as in the case of TBT and TIT, or is found to become less intense and shifted to lower frequencies. Similarly ν (C=S) band (~ 720 nm) is also reduced in intensity. Thus, the ir spectra of these complexes indicate that the pyridyl nitrogen and thiocarbonyl sulfur act as donors, and the complex is 4-coordinated.

TABLE II

Ultraviolet spectral data on the Cu (I) and Zn (II) complexes

Compound	Solvent	λ_{max} (nm)	E_{max} ($1\ mol^{-1} cm^{-1}$)
CuCl $(PCT)_2$	Chloroform	273	36160
		313	39120
CuCl $(PBT)_2$	do.	271	40790
		317	47400
CuCl $(PIT)_2$	do.	276	34020
		317	42240
CuCl $(TCT)_2$	do.	261	30660
		313	37480
CuCl $(TBT)_2$	do.	262	31960
		314	40190
CuCl $(TIT)_2$	do.	276	31700
		319	42030
Zn $(PCT)_2$ $(ClO_4)_2$	DMF	276	24730
		311	22650
Zn $(PBT)_2$ $(ClO_4)_2$	do.	276	22810
		309	20120
Zn $(PIT)_2$ $(ClO_4)_2$	do.	282	20250
		314	18950
Zn $(TCT)_2$ $(ClO_4)_2$	do.	274	21310
		311	19270
Zn $(TBT)_2$ $(ClO_4)_2$	do.	278	28110
		309	24890
Zn $(TIT)_2$ $(ClO_4)_2$	do.	281	30900
		315	26720

DMF = Dimethylformamide.

TABLE III

Infrared spectral peaks on the Cu (I) and Zn (II) Complexes (in nujol-mull)

Compound	ν (C=C + C=N) (cm^{-1})	ν (N–C–N + C=S) (cm^{-1})	ν (C=S) (cm^{-1})
PCT	1598 s	1070 m	720 m
CuCl $(PCT)_2$	1600 s	1065 w	715 m
Zn $(PCT)_2$ $(ClO_4)_2$	1600 s	1025 w	715 sh
PBT	1595 s	1070 m	720 m
CuCl $(PBT)_2$	1598 s	1065 vw	710 w,b
Zn $(PBT)_2$ $(ClO_4)_2$	1590 s	1017 vw	715 vw
PIT	1592 s	1077 m	716 m
CuCl $(PIT)_2$	1585 s	1070 m	705 vw
Zn $(PIT)_2$ $(ClO_4)_2$	1590 s	1015 vw	710 vw
TCT	1600 s	1042 m	705 m
CuCl $(TCT)_2$	1598 s	1045 w	710 m
Zn $(TCT)_2$ $(ClO_4)_2$	1600 s	1045 vw	700 w
TBT	1595 s	1042 m	705 m
CuCl $(TBT)_2$	1598 vs	1040 w	705 w
Zn $(TBT)_2$ $(ClO_4)_2$	1592 s	..	700 w
TIT	1590 s,b	1077 m	703 m
CuCl $(TIT)_2$	1595 s	1077 m	715 w
Zn $(TIT)_2$ $(ClO_4)_2$	1590 s,b	..	700 w

Note : w = weak, v = very, sh = shoulder, m = medium, s = strong and b = broad.

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1. Dutta Ahmed, A. and Mandal, P. K., *J. Inorg. Nucl. Chem.*, 1967, **29**, 2347.
2. Banerjea, D. and Singh, I. P., *Ind. J. Chem.*, 1968, **6**, 34.
3. Krishnaswamy, N. and Bhargava, H. D., *Ibid.*, 1969, **7**, 710.
4. Kashyap, B. C., Taneja, A. D. and Banerji, S. K., *J. Inorg. Nucl. Chem.*, 1975, **37**, 612.
5. Schumacker, J. C., *Perchlorates—Their Properties, Manufacture and Uses*, ACS Monographs, Reinhold, New York, 1960, p. 107.
6. Vogel, A. I., *A Text Book of Quantitative Analysis*, Longmans Green and Co., London, 1943.
7. Pearson, R. G., *J. Amer. Chem. Soc.*, 1963, **85**, 3533.
8. Ahrland, S., Chatt, J. and Davies, N. R., *Quart. Rev.*, 1958, **12**, 265.
9. Hathaway, B. J. and Underhill, A. E., *J. Chem. Soc.*, 1961, p. 3091.

COLORIMETRIC STUDIES ON THE REACTION OF U (VI) AND Mo (VI) WITH 3-5-DICHLORO-2-HYDROXYACETOPHENONE AND ITS OXIME

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ABSTRACT

Chelate formation of uranium and molybdenum with 3, 5-dichloro-2-hydroxyacetophenone and its oxime have been studied. The mole-ratio (M : L) was found 1 : 2. The stability constant of these metal chelates at different ionic strength and their dieclectric constants are calculated.

THE *o*-hydroxyaldoximes are well known as analytical reagents. Ephraim[1] was first to investigate salicylaldoxime as a reagent for detection and determination of copper. His work was extended by number of other workers. Poddar[2] studied *o*-hydroxyacetophenone oxime as an analytical reagent and found that this reagent has definite advantages over salicylaldoxime due to the greater thermal stability and increased insolubility of metal complexes.

The formation of complexes of metals with 3, 5-dichloro-2-hydroxyacetophenone oxime have been recently reported by Gupta and Lal[3-4]. The present note describes the colour reactions of uranyl and molybdate ions with 3, 5-dichloro-2-hydroxyacetophenone (HDCA) and 3, 5-dichloro-2-hydroxyacetophenone oxime (HDCAO).

EXPERIMENTAL

Stock solutions of uranyl nitrate and ammonium molybdate (B.D.H.), (AnalaR) were prepared in distilled water and their strength determined by usual methods. All other chemicals used were of B.D.H. (A.R.) grade. Buffers of ammonium hydroxide and acetic acid were used to adjust the pH of the solution. The reagents HDCA and HDCAO were prepared as reported earlier[3]. Absorbance and pH measurements were carried out with systronic spectrocolorimeter (type 102) (range 400–700 nm) and pH meter (type 322) respectively.

RESULTS

HDCAO forms instantaneously an orange-yellow coloured, soluble complex with uranyl and molybdate ion. The complex is stable at room temperature. It was observed that the colour of the molybdenum (VI) complex was stable upto 45° C. However, the solution regained its original colour intensity on cooling to room temperature. HDCA also gives a orange-yellow soluble complex with uranyl ion.

The nature of the complexes was studied by the method suggested by Vosburgh and Cooper[5] and found that only one complex was formed under the condition of study in all the systems. The method of continuous variation[6], slope-ratio[7], mole-ratio[8] methods were used in order to establish the molar ratio of metal-ligand. The physical constants of the complexes were calculated using the well known spectrophotometric methods. The results obtained are recorded in Table I.

TABLE I

Physical constant of UO_2 (II) and MoO_2 (II) complexes with HDCAO and HDCA

Metal ion	Colour	λ_{max} (nm)	pH	M : L	Beer's law range ppm	Stability constant (μ = 0·1 M)	Sensitivity molar extinction coefficient
			Complexation with HDCAO				
UO_2 (II)	orange-yellow	400	7·5–8·0 (7·5)	1 : 2	20–110	5·50	0·55 μg U/cm² 430
			Complexation with HDCAO				
UO_2 (II)	orange-yellow	400	6·0–8·5 (7·5)	1 : 2	17–90	6·39	0·33 μg U/cm² 730
MoO_2 (II)	orange-yellow	400	2·5–3·5 (3·0)	1 : 2	2–18	7·28	0·19 μg Mo/cm² 510

(Parentheses value refer to the pH of study).

To study the effect of pH on the complex formation, a large number of solutions were prepared containing the metal and the ligand, the latter being in large excess (15–25 times) in order to obtain maximum colour developments. The spectral readings were recorded at λ_{max} of the complex at various pH values.

Evaluation of Stability Constant :

The stability of the chelates have been calculated by mole-ratio plot, using the relations $K = (1 - \alpha)/4c^2\alpha^3$ and $\alpha = (Em\text{–}Es)/Em$ where Em, Es, C and α have the usual significance. With a view to obtaining the thermodynamic stability constant (K) of the chelates, the K at different ionic strength (μ) ($NaClO_4$) studied at suitable pH Table II.

TABLE II

Stability constant of UO_2 (II) and MoO_2 chelates at various ionic strengths (μ)

(Solvent, 75% v/v ethanol-water)

Ionic strength (μ)	log K for UO_2 $(L_1)_2$	log K for UO_2 $(L_2)_2$	log K for MoO_2 $(L_2)_2$
0·15	5·45	6·30	7·20
0·10	5·50	6·39	7·28
0·05	5·53	6·43	7·34
0·02	5·58	6·52	7·41
0·00	5·62*	6·60*	7·90*

* By Extrapolation method.
L_2 = HCDAO and L_1 = HDCA.

By the method of extrapolation using the log K values, the thermodynamic stability constant and free energy (△ F°) at zero ionic strength has also been calculated.

The △ F° was found to be 7·73, 9·07 and 10·86 KCal/mole at 27° C, for the chelates $UO_2(L_1)_2$, $UO_2(L_2)_2$ and $MoO_2(L_2)_2$ respectively. HL_1 and HL_2 are the legands of HDCA and HDCAO respectively.

In order to study the effect of dielectric constant on the stability constant of the chelates, several different percentage of ethanol-water solutions were used. The results are tabulated in Table III,

TABLE III

Stability constants of UO_2 (II) and MoO_2 (II) chelates in Different Ethanol water solvent mixtures

(Ionic strength 0·1 M)

Ethanol (%)	Dielectric constant*	log K for UO_2 $(L_1)_2$	log K for UO_2 $(L_2)_2$	log K for MoO_2 $(L_2)_2$
44·00	51·8C	5·35	6·16	7·04
54·0	46·20	5·40	6·23	7·14
65·0	40·00	5·44	6·32	7·22
75·0	34·00	55·0	6·39	7·27

* Akerlof, G., *J. Am. Chem. Soc.*, 1932, **54,** 4125.

DISCUSSION

It is evident from Table I, that metals form orange-yellow colour chelates with HDCA and HDCAO in the stoichiometric ratio of M : L is 1 : 2.

From Table II it can be noted that stability constant of the chelates of molydenum and uranium increases with decreasing ionic strength. The stability constant of the chelates at different percentage of ethanol reveals that the stability constant slightly increases with the decrease in the dielectric constant (ϵ) (Table III) of the media, although the nature of the spectrum does not change with the organic solvent. Optical density increase as the percentage of alcohol increase. In such cases, it is expected that the dielectric constant of the media effect the stability constant of chelate to a great extent. It is noteworthy that an approximate linear relationship is obtained when log K is plotted against $1/\epsilon$.

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1. Ephraim, F., *Ber. dt. chem. Ges.*, 1930, 63, 1928 ; 1931, 64, 1210, 1215.
2. Poddar, S. N., *Z. analyt. Chem.*, 1957, 154, 254 ; 1957, 155, 327 ; *Analytica Chim. Acta*, 1963, 28, 586 ; *J. Indian Chem. Soc.*, 1963, 40, 706 ; *Indian J. Appl. Chem.*, 1963, 26, 153.
3. Gupta, S. P., Srivastava, S. K. and Lal, K., *Indian J. Chem.*, 1975, 13, 297.
4. Lal, K. and Gupta, S. P., *Curr. Sci.*, 1975, 44, 178.
5. Vosburgh, W. C. and Cooper, G. R., *J. Am. Chem. Soc.*, 1941, 63, 437.
6. Job, P., *Ann. Chim.*, 1928, 9, 113.
7. Harrey, A. E. and Manning, D. L., *J. Am. Chem. Soc.*, 1950, 72, 4488.
8. Yoe, J. H. and Jones, A. L., *Ind. Eng. Anal. Ed.*, 1944, **16,** 111,

THE EPARCHAEAN UNCONFORMITY AT TIRUMALAI—A STUDY

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ABSTRACT

The Tirumalai Hill Range, with the scarp topography and with the two ghat roads dissecting it, presents several interesting geological features, one of which is the unconformable contact between the granite basement over which lies the Cuddapah quartzite. The bedded quartzite, with very low dips, forms escarpments, whereas the granite beneath has flaring slopes. As the quartzite is deposited over the granite domes and dolerite ridges, the latter appear to project themselves into the former. It is stressed here that the contact between the granite and quartzite is depositional and not intrusive. The presence of granite pebbles in basal conglomerate is of great significance, as such occurrences are rare. The basement granite has diversely oriented joints and is also traversed by dykes. The igneous activity and the stresses that promoted the formation of fracture system in the granite had ceased before the epeirogenic movement responsible for the deposition of Cuddapah sediments and its uplift intervened.

INTRODUCTION

THE steep scarp of the Tirumalai Hill Range, rising to a height of over 3,000 ft. from the plains of an average height of 500 ft. at the Tirupati township (India), presents a remarkable evidence of an unconformity in its topographic, structural, and denudational features. This unconformity is the Eparchaean unconformity—a universal phenomenon—not the least well-represented in the Cuddapah basin, of which the Tirumalai sediments form a part. This statigraphic break occurs between the crystalline schists, gneisses, and the granites of the Archaean and the subsequent group, called the Puranas (Proterozoic or late Precambrian). In his admirable portrayal of this gap in the geological record, Pascoe (1939) pictures a prolonged period of denudation to which the Archaeans were exposed, and thus to the enormous time interval prior to the deposition on their denuded surface of the later sediments.

THE TIRUMALAI UNCONFORMITY

Topographic Expression

Dealing with the topographic features first, one can see, looking north from the Tirupati side, a flat, or gently northerly dipping quartzite exposed in the vertical cliffs forming ascarpment to the south. The quartzite is yellowish brown and is barren of vegetation. Beneath the quartzite lies the granite covered by a thin layer of residual soil that supports a sparse vegetation. The sudden appearance of vegetation from the unconformable junction downwards is very striking. The granite massif from its unconformable contact with the quartzite, has a flaring slope in bells and disappears in the plains of Tirupati under a thick cover of gravelly soil to rise again farther south as domes and tors. Along its flaring slopes could be seen, here and there, dome-like projections of bare granite with exfoliated surface. This topographic discordance between the granite and the underlying quartzite is a result of structural and mineralogical differences between the two rocks which cause differential erosion to take place. The granite is a coarse-grained porphyritic granite. The phenocrysts of potash feldspars, have random orientation. Neither foliation nor lineation is present, despite the fact that biotite is the mineral present as a mafic constituent. The porphyritic granite has thus a mesoscopic isotropic fabric. These features together with the fact that the granite is coarse-grained promote more rapid disintegration (Lahee, 1961, p. 26). This, together with concentric weathering and exfoliation, produces sphericity in the granite to form dome-shaped masses. The granites are traversed by joints which have diverse orientation. These diversely oriented joints promote removal of exfoliated sheets during the process of weathering to produce domical surfaces. The overlying quartzites have dip of 10° to 13° and, at places, are almost horizontal. They are characterised by bedding joints and the master joints which stand vertical. The weathering of quartzite takes place, perforce, by the mechanical process of dislodgement of blocks by separation along joint planes. Thus, are produced the steep, vertical scarps in quartzite. The quartzite is fine-grained, homogeneous and, hence, produces sharp edges and corners. There is a pointed reference to this type of weathering by Lahee (1961, p. 388).

The erosion surface on which the Tirumalai quartzite is deposited is not a flat surface; it is a highly irregular undulating surface. Along the south-facing, east-west trending scarp of Tirumalai, the granite has a number of rises and falls. The rises between two depressions appear to protrude into the flat, horizontally bedded quartzite to give an erroneous impression of the granite to be an

intrusive body. This impression is much more misleading in the case of dolerites which occur as long ridges and stand out in bold relief above the granite outcrops and meet the quartzites in protuberances. Vaidyanadhan (1957) refers to a dolerite dyke at 9/4 miles along the old ghat road, as having cut through not only the granite but also the overlying quartzite, and says, that this intrusive is obviously younger than the quartzose sandstone. On close examination, this dolerite was found to have a domical contact with the quartzite and is never found cutting across the quartzite. The ghat road has exposed the quartzite in a vertical cliff. The quartzite is horizontal, thickly bedded, and overlies the eroded domical surface of the dolerite as a thick cover.

The Depositional versus *Intrusive Contact*

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The erosion surface on which the Tirumalai quartzite is deposited is not a flat surface ; it is a highly irregular undulating surface. Along the south-facing, east-west trending scarp of Tirumalai, the granite has a number of rises and falls. The rises between two depressions appear to protrude into the flat, horizontally bedded quartzite to give an erroneous impression of the granite to be an

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in the conglomerate is of significance as such occurrences are rare, and calls for the examination of the Cuddapah basin, not only at its fringes but also in the interior of the basin (as at the Venkateswara shrine) to reveal whether any variation in the type of pebbles occurs from place to place. The Cuddapah sediments were originally horizontally bedded but were tilted to attain a dip of 10° to 13° due to epirogenic uplift.

1. Lahee, F. H., *Field Geology*, McGraw-Hill Book Co., Inc., 1961.
2. Pascoe, E. H., *A Manual of Geology of India and Burma*, Government of India, 1950.
3. Vaidyanadhan, R., "Significance of certain sedimentary structure in Cuddapah sandstones," *Quart. Jour. Geol. Min. Met. Soc. India*, 1957, 29 (4), 191.

NODULATION PATTERN IN GROUNDNUT (*ARACHIS HYPOGAEA* LINN.) AS INFLUENCED BY VARYING LEVELS OF PLANT DENSITY NITROGEN AND PHOSPHORUS*

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ABSTRACT

Nodulation in groundnut was initiated at flowering, profuse at pod formation and continued upto 85 days. Nodules started degenerating after 85 days. Highest plant density treatment (2,00,000 plants/ha) was found to produce significantly less number and small sized nodules per plant, only at latter stages of nodulation in both the seasons. The medium (1,50,000 plants/ha) and low (1,00,000 plants/ha) densities were found at par. The retarding effect of nitrogen, on nodulation applied either at 20 or 40 kg. N/ha over the control was observed at active growth phase in both the seasons. However, in the initial stage of nodulation, and at 85 days, the effect of nitrogen was not significant. The positive effect of phosphorus in increasing the volume of nodules was evident in the initial stage of nodulation. The dose of 60 kg P_2O_5/ha was found effective over 20 and 40 kg P_2O_5/ha, especially under low population. The lower doses of phosphorus were not effective.

1. INTRODUCTION

It is essential to know the pattern of nodulation in order to harness biological nitrogen fixation effectively for increased groundnut production through root nodule bacteria (*Rhizobum leguminosarum*) by symbiosis especially under present day fertilizer constraint. Many internal and external factors influence the extent of nodulation. Supply of nutrients and density of plant population are important factors controlling nodulation under average farming conditions. In this communication we describe the results of field experiments on the pattern of nodulation in groundnut crop grown under varying levels of plant density, nitrogen and phosphorus.

2. METHODS AND MATERIAL

A field experiment was conducted on groundnut crop, variety AK 12–24 at the Agricultural College Farm, Akola in *Kharif* seasons of 1971 and 1972. The three factor experiment was conducted in split plot design. Nine treatment combinations formed by combining 3 rates of planting one lakh S_1, one and half lakhs S_2, and two lakhs S_3, of plants per ha with 3 levels of nitrogen 0 (N_0), 20 (N_1) and 40 (N_2) kg N/ha were tried as the main plot treatments. The four levels of phosphorus, 0 (P_0), 20 (P_1), 40 (P_2) and 60 (P_3) Kg P_2O_5/ha were superimposed as subplot treatments. The set of 36 treatment combinations, was replicated three times. Each experimental unit was 9 m × 7·2 m, and 7·2 m × 5·4 m in gross and net plot sizes respectively. The site and randomization were changed in second season. Five plants at each stage were selected at random from each net plot and uprooted carefully for studies on number and volume (cc) of nodules per plant. Nodulation studies were taken up in second and third months. Nodules started degenerating in fourth month, hence data for fourth month are not recorded.

3. EXPERIMENTAL FINDINGS AND DISCUSSION

Data on mean number of nodules and their volume in cc per plant collected at various stages in both the seasons are presented in Table I.

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* Contribution from the Department of Agronomy, Punjabrao Krishi Vidyapeeth, Akola.

TABLE I

Mean number (N) and volume (V) of nodules in cc per plant

	I Season								II Season							
Days after sowing	40		55		70		85		36		51		64		79	
	N	V	N	V	N	V	N	V	N	V	N	V	N	V	N	
S_1	27·0	0·049	53·2	0·19	79·0	0·26	107·9	0·40	28·7	0·068	52·8	0·20	81·6	0·27	98·7	0·40
S_2	27·7	0·052	51·4	0·20	93·0	0·32	117·9	0·48	28·7	0·062	54·5	0·21	95·1	0·33	119·6	0·49
S_3	28·9	0·078	59·0	0·21	97·0	0·33	117·1	0·50	30·2	0·059	60·4	0·22	98·8	0·34	129·8	0·51
S.Em ±	0·79	0·004	2·95	0·01	4·9	0·02	8·6	0·04	1·1	0·005	3·2	0·01	4·8	0·02	8·4	0·04
C.D.	NS	NS	NS	NS	14·5	0·05	NS	NS	NS	NS	NS	NS	14·2	0·05	25·0	NS
N_0	29·6	0·07	62·4	0·23	96·4	0·33	122·1	0·49	30·7	0·063	64·2	0·24	98·0	0·34	123·8	0·48
N_1	28·0	0·05	52·4	0·21	89·9	0·30	116·4	0·47	29·2	0·060	54·0	0·22	91·7	0·31	118·5	0·50
N_2	26·1	0·05	48·8	0·17	82·6	0·29	104·3	0·42	27·8	0·065	49·4	0·18	85·9	0·30	105·9	0·43
S.Em ±	0·79	0·004	2·95	0·01	4·9	0·02	8·6	0·04	1·09	0·005	3·2	0·01	4·8	0·02	8·4	0·04
C.D.	2·36	NS	8·82	0·03	NS	NS	NS	NS	NS	NS	9·5	0·03	NS	NS	NS	NS
P_0	26·3	0·05	58·7	0·20	84·9	0·28	110·6	0·46	28·2	0·059	52·5	0·20	86·7	0·30	112·0	0·46
P_1	28·1	0·05	51·6	0·19	87·7	0·30	115·4	0·46	28·7	0·059	53·6	0·21	90·4	0·30	117·4	0·47
P_2	27·3	0·05	56·3	0·21	88·1	0·31	115·4	0·45	29·0	0·059	58·0	0·22	90·6	0·32	116·5	0·46
P_3	29·8	0·06	52·3	0·20	97·9	0·34	116·0	0·47	31·1	0·074	59·4	0·21	100·0	0·34	117·7	0·48
S.Em ±	1·16	0·004	3·9	0·01	5·0	0·02	6·4	0·02	1·8	0·004	4·0	0·01	5·1	0·02	6·8	0·02
C.D.	NS	0·010	NS	NS	NS	NS	NS	NS	NS	0·012	NS	NS	NS	NS	NS	NS
Mean	27·9	0·053	54·5	0·20	89·6	0·31	114·3	0·46	29·2	0·063	55·9	0·21	91·9	0·32	116·0	0·47

3.1 : *Number of Nodules*

Table I indicated that the nodulation in groundnut was initiated at flowering, profuse at pod formation and continued upto 85 days. Jaubert[8] found initiation of nodules three weeks after sowing, Masefield[10,11] at flowering ; while Seshadri[13] reported the initiation at the end of first fortnight. Continued nodulation upto harvest was reported by Seshadri[13], but Bharad[3] observed degeneration after 66 days.

The effect of planting rate on number of nodules per plant was not significant at initial stages of nodulation. But at later stages (70 days in first season and 64 and 79 days in second season), the treatment of low planting rate was found to bear significantly more nodules per plant than that of high rate of planting (because of limited growth due to competition). However, the medium (1,50,000 plants/ha) and low (1,00,000 plants/ha) densities were found at par.

The effect of nitrogen on the number of nodules per plant was not evident at the initial as well as the later stages of nodulation ; but it was evident at active growth phase (40 and 55 days in first season, 51 days in second season). The dose of 40 kg N/ha was found to reduce significantly the number of nodules per plant as compared to the control. It is due to the fact that as the nitrogen need of the plants was fulfilled through increased supply of nitrogen from fertilizer, the plants tended to bear less nodules. Retarding effect of nitrogen fertilizer on nodulation was also observed by Ali Mohammad *et al.*[1], Bouger[4], and Gopalkrishnan and Nagarajan[7]. However, the necessity of small amount of nitrogen was noted by Ludwig and Alison[9], Burkhat and Collins[6], Jaubert[8], Bunting[5] and Seeger[12]. But, Thornton[14] observed that nodulation was not affected by nitrogen fertilization.

The effect of phosphorus influencing the number of nodules per plant was not evident at all stages in both the seasons. Thornton[14] and Bharad[3] could not notice the beneficial effect of phosphorus on nodulation ; whereas Baltan[2], Bouger[4] and Gopalkrishnan and Nagarajan[7] could notice it.

The effect of S × N interaction at 40 days in the first season and 36 days in the second season, and that of S × P interaction at 40 days in the first season, were found significant. The corresponding data are presented in Tables II and III respectively. The effects of other interactions were not significant.

Table II indicated that the reciprocal effect of density on nodulation was evident only in control plots due to the limited growth of plants in high density low nitrogen plots. Similarly, the retarding effect of nitrogen on nodulation was observed only at the low density where there was no competition for fertilizer nitrogen.

TABLE II

Mean number of nodules per plant as influenced by S × N at 40 days in I season and 36 days in II season

Treatments	N_0	N_1	N_2	S.E.m (SN)	C.D. (SN)
S_1	27·05 (28·33)	26·85 (28·16)	26·90 (30·09)	1·370 (1·893	4·096 (5·660)
S_2	29·94 (29·52)	27·90 (28·52)	25·32 (27·97)		
S_3	31·40 (34·16)	29·16 (30·89)	26·20 (25·46)		

Figures in Parenthesis correspond to II season.

TABLE III

Mean number of nodules per plant as influenced by S × P interaction at 40 days in I season

Treatments	P_0	P_1	P_2	P_3	S.E. (SP)	C.D. (SP)
S_1	25·27	27·53	26·85	27·90	2·010	5·570
S_2	26·33	28·15	27·32	29·08		
S_3	26·85	28·53	27·73	32·57		

It is observed from Table III that nodulation was enhanced under the condition of high phosphorus (60 kg P_2O_5/ha) and low rate of planting (1,00,000 plants/ha). At lower doses of phosphorus and high plant density, however, such beneficial effect was not evident.

3.2 : *Volume of Nodules*

Table I would indicate that the effect of plant density, nitrogen and phosphorus on volume of nodules per plant were similar to those observed in case of number of nodules per plant except that the main effect of phosphorus in producing bigger sized nodules was prominent in initial stage. The dose of 60 kg P_2O_5/ha was found sufficient.

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1. Ali Mohammad Alam, Alam Zafar and Khanna, K. L., *Agr. and Livestock in India*, 1932, 3, 91.
2. Baltan, E. T., *Va. Agr. Espt. Sta. Bull.*, 1922, 229.
3. Bharad, G. M., *A Thesis Submitted to Nagpur Univ. for M.Sc.* (*Agri.*), 1964, p. 1.

4. Bouger, S., *Agron. Trop.*, 1949, 4, 229 ; *Noted from Indian Oilseeds J.*, 8 (3), 222.
5. Bunting, A. H., *Nature*, 1956, 178, 1102.
6. Burkhart, L. and Collins. E. R., *Soil Sci. Soc. Amer. Proc.*, 1942, 6, 272.
7. Gopalkrishnan, S. and Nagarajan, S. S., *Indian Oilseeds J.*, 1958, 2, 5.
8. Jaubert, P., *Bull. Agron. Fr. D'out Met.*, 1952, 7, 141 ; *Noted from Fld. Crops Abstr.*, 1958, 11, 1 and 2.
9. Ludwig, C. A. and Allison, F. E., *Agron. J. Amer. Soc.*, 1935, 27, 895.
10. Masefield, G. B., *Emp. J. Expt. Agr.*, 1952, 20 (79), 175.
11. —, *Ibid.*, 1957, 25 (98), 175.
12. Seeger, J. R., *Bull. Inst. Agron. Gembl.*, 1961, 29 (2), 197.
13. Seshadri, C. R., *Groundnut-Monograph. I.C.O.C.*, Himayatnagar, Hyderabad, Deccan-1, 1962.
14. Thornton, I., *Oleagineax*, 1963, 18 (2), 781 ; Noted from *Trop. Abstr.*, 1964, 9 (4), 239.

FIRST INDIAN GEOPHYTOLOGICAL CONFERENCE

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THE Paleobotanical Society, organised the First Indian Geophytological Conference in Lucknow from 21st to 24th December, 1975. The complexion of the Conference was the combination of Geological Sciences, Biological Sciences (including Paleobiological Sciences) and Oil Sciences. Although the Conference had the title Geophytological Conference, it was more than paleobotanical in its deliberations. The Conference was held in 8 sessions.

Two sessions were devoted to plant morphology and taxonomy where twentythree papers were presented highlighting the importance of the pollen morphological studies of the tri-colporate, the ontogony of stomata of some members of Apocynaceae (Trivedi and Upadyaya), the oil-bodies in South Indian liverwort (Udar and Nath) and the genetic engineering for certain ornamentals (Raghuvanshi and others).

Two sessions were devoted to Paleobotany and Phylogeny where twenty papers were presented on the following subjects : (1) A new genus from the Deccan Intertrappean beds of Madhya Pradesh (B. S. Trivedi and others), (2) A new genus and species based on the study of wood material (K. S. Patil), (3) The new taxa, of *Chara, Agaricus* and *Nitellites,* from the Intertrappean beds of different regions (T. V. Sivarudrappa, T. K. Trivedi, S. B. Batia and A. R. Kulkarni), (4) New species *Ottocaria, Palmoxylon* and *Ptilophyllum* (Manju Banerjee, S. S. Berlinge, S. A. Paradkar, Sukdev and Zeba Banu). The papers dealing with fossil algae were presented by A. K. Pal and others on the assemblage of undoubted Permian age from Garhwal Himalaya. Only one paper dealing with the Pollen was presented by S. K. Bakshi and Urmila Deb. This paper covered the Cenozoic sequence of Assam and Bengal regions and the morphological evolutionary trends of two genera.

One session was devoted to stratigraphy at which thirteen papers were presented. The papers covered stratigraphic aspects of Algal Stromatolites of Vindhyans of Western Rajasthan (G. Barman and K. K. Varma), spore-pollen assemblage from Iran and Kobra Coal Fields (D. C. Bharadwaj and others), Paleo-stratigraphic zonation of the coal bearing Horizon of Godavari (A. A. Mohiz and N. Ramana Rao), Palino-stratigraphy of the Mikir Formation (S. C. D. Sah and others), Paleontological demarcation of the Subathu and Dagshai sediments of Simla Hills (H. P. Singh and others) and Geologic range of Fossil Algae from the Mesozoic rocks of Trichinopoly District of Tamil Nadu, South India (S. Sambe Gowda). This paper dealt with the stratigraphic range of leading genera and also the genus *Corallina.* The fossil algae in this region could be used for paleoecological interpretations. The paper by M. N. Vishwanathaiah and others on the stratigraphic position of Badami group dealt with the acritarchs groupable into three distinct assemblages ranging in age from Ordovician to Silurian. The paper by D. C. Bharadwaj and A. Dwivedi dealt with the statistical approach to the circumscription of species in spore disperse.

The Conference was also concerned with the plant ecology and geography. These papers were concerned with geographical and ecological aspects of the living as well as the fossil plants. Paper by Ramanujum was concerned with the palynological approach in regard to the study of palaeoecology and palaeogeography of the plant kingdom represented by Warkalli deposits of Kerala. Paper by Thothathri dealt with phytogeographic studies of Andaman and Nicobal regions. Paper by Patnaik and Sharma concerned itself with preliminary studies of economically important plants of the Ganjam District in Orissa State.

Session VIII was devoted to sedimentology and biopetrology with eight papers. One paper by Navale dealt with the Biopetrology of Kargali seam of the East Bokaro Coal Field in Bihar, the rest of the papers were concerned with the sedimentological studies of rocks of some regions in the Central and Northern parts of India. Paper by Basumallick and Sharma was concerned with the discovery of Glauconites from Eocene carbonates of Assam region. The paper by Arabindha Ghosh on the overgrowth in Albite and K-Feldspar, as found in the fresh-water Lameta beds, was of interest because of his reasoning that the marine conditions need not necessarily be ideal for Na+ and K+ metasomatism. A paper by A. R. Bhattacharya was concerned with Zircon and Tourmaline, from the Precambrian arenaceous rock units of distinct Lithostratigraphic units. He has shown that the shape and size of these two minerals can be used as distinct criteria for recognizing the differences in apparently similar-looking rock units.

Of the two symposia, one was concerned with the soil plant relationship and the other with the climatic vicissitudes in India during the Gondwana times. In the former symposium there were as many as twentyfive papers, all dealing with the mineralogical influence on the growth of plants and their distribution pattern. The latter symposium had eight papers dealing with the climatic vicissitudes of the Gondwana Era in India. The discovery of lowest Gondwana beds, equivalents of Talchirs, consisting of unsorted boulders and khakhi shales in the Chingelpet District of Tamil Nadu, discovered by the workers of G.S.I. was not, unfortunately, a part of the review presented at the session by one of the workers of the G.S.I.

The Conference was inaugurated by Vice-Chancellor Kaul who dealt with the palaeogeographic distribution of plants of the South Asian and Australasian regions. The Presidential address by Prof. R. C. Mishra dealt with the application of geobiological techniques in the exploration for economically important mineral deposits. The author of this report hopes that the first geophytological conference would be a happy augury for many more in the years to come.

INTERNATIONAL SYMPOSIUM ON 'USE OF NON-HUMAN PRIMATES IN BIOMEDICAL RESEARCH'

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RECOGNIZING the importance of non-human primates in biomedical research to the overall welfare of mankind, the Indian National Science Academy sponsored an international symposium on the 'Use of non-human primates in biomedical research'. The objectives of the
sensus on the need for proper and rational utiliza-
symposium were : (1) to evolve an international con-
tion of non-human primates to avoid depletion of wild populations by the growing needs of research, (2) to direct efforts towards formulation of scientifically sound programmes of management and conservation of non-human primates as a valuable natural resource.

The symposium was held in New Delhi from November 3–8, 1975 and was attended by 120 leading biomedical scientists from 13 countries, representatives from the World Health Organization and from all research institutions and agencies in India. Forty-two papers were presented, dealing with the use of non-human primates in areas of biomedical research directly relevant to human health and welface, namely, reproductive biology, contraceptive technology, nutrition, neurobiology, surgery, infectious diseases, immunology, psychopharmacology, toxicology, pharmacology and the study of non-human primates in the wild and their conservations.

The discussions focussed attention on issues of vital importance, namely :

(*a*) relevance of using non-human primate models in biomedical research and other biomedical endeavours.

(*b*) Consideration of steps to be taken to conserve non-human primate populations which represent a valuable natural resource.

(*c*) specific recommendations for judicious and restrained use of non-human primates in biomedical research.

The international community of scientists gathered at the symposium reached the following conclusions :

The most dramatic illustration of the importance of non-human primates in medical research is poliomyelitis vaccine which was produced using polio viruses grown in tissue cultures of monkey kidney cells and is tested in monkeys. Many other examples were cited in the fields of infectious and parasitic diseases, reproductive biology and contraceptive technology, etc. Biomedical scientists are aware of the increasing pressures on numerous primate animal species caused by the growth of human populations and agriculture. Non-human primate species should be protected by appropriate conservation measures. Review committees and granting agencies should review research projects so as to avoid wasteful use of primate animals. Animal husbandry methods should be adopted for efficient breeding of non-human primates to supplement those caught in the wild and to provide specific-pathogen-free animals of known parentage. Judicious cropping of the more abundant species and management practices may be expected to improve the total supply.

Biomedical research can, in fact, help to conserve monkeys by developing specific programmes of conservation and breeding. Multiple utilization of individual animals is accepted and advocated by this group. Laboratory scientists are not in conflict with conservation and policies and endorse the need to preserve animals in the wild. Management of non-human primates in the wild and judicious cropping is preferable to prohibition of their use in biomedical research. The scientists noted with satisfaction that the regulations for capture, transport, caging and husbandry of primates formulated and enforced by the Indian Standards Institution, had improved considerably not only the quality of animals now available for research, but had also decreased the wastage of animals during transport. Endangered or rare species of primates (as listed in the I.U.C.N. Red Data Book) should not be harvested from the wild for biomedical research. The study of the natural history of these species should be intensified to ensure their survival.

The group made the following specific recommendations for improving primate research facilities in India :

Standard guidelines regarding husbandry, quarantine, caging, feeding, breeding, etc., should be made available to the scientists using non-human primates in biomedical research. Priority should be given to primate field studies on population and on diseases occurring in the wild. Proper conservation and management of India's valuable primate resources require extensive study of the ecology, distribution, abundance, population dynamics, habitat utilization and social behaviour of the species occurring in the wild. Such studies will lead to formulation of policies for management of wild populations of primate animals to ensure conservation of the species and to provide adequate supplies for biomedical research. The group recommended that the Government of India give active consideration to the implementation of the following programmes in a phased manner :

Establishment of ranches (farms) under natural conditions from which monkeys could be periodically cropped, quarantined and supplied to investigators. The programme could be expanded to meet world-wide needs.

The establishment of one or more primate research institutes should be considered by the Government to provide facilities for visiting scientists from other institutions and for conduct of research in primate biology. These facilities may form the nucleus of an International Primate Research Centre on the lines of similar international research institutes.

INDIAN SOCIETY OF DEVELOPMENTAL BIOLOGISTS

THE above society came into existence on 4th January 1976, at a meeting held during the 63rd Session of Indian Science Congress Association at Waltair. The general body meeting elected Professor Har Swarup, Head of the Zoology Department, Vikram University, Ujjain, as President and Dr. Suresh C. Goel, Reader in Developmental Biology, University of Poona, Poona, as Secretary-Treasurer of the Ad–Hoc Governing Board of the Society. Persons interested in promoting the cause of developmental biology in India are requested to contact Dr. Goel, for further details.

LETTERS TO THE EDITOR

EXTRACTIVE SPECTROPHOTOMETRIC METHOD FOR THE DETERMINATION OF BISMUTH WITH TETRAPHENYLARSONIUM CHLORIDE

Introduction

THE use of tetraphenylarsonium chloride as an analytical reagent has long been known[1-3]. The complex halides of many metals and the anions, *viz.*, ClO_4^-, IO_4^- and ReO_4^- form insoluble precipitates with the reagent. The pricipitates have been used in the gravimetric analysis. Based on the solubility of some of these precipitates in chloroform, anions like ReO_4^-, TcO_4^- and MnO_4^- have been extracted into chloroform[4]. Similar ion pairs of the reagent with anions such as chloro and thiocyanato complexes of bismuth, cadmium, iridium (IV)[5], niobium[6], niobium and titanium[7], palladium[8], zinc, tin (IV) and gold[9] have been used in the extractive spectrophotometric method. A similar reagent, tetraphenylphosphonium bromide has been used in the extractive photometric determination of bismuth[10]. Bismuth, in presence of potassium iodide, was found to give a similar precipitate with tetraphenylarsonium chloride having higher solubility in 1, 2-dichloroethane than in chloroform. In the present method, the extractive spectrophotometric determination of bismuth as tetraphenylarsonium tetraiodobismuthate in 1, 2-dichloroethane was investigated.

Experimental

Absorbance measurements were made with a Beckmann DK-2 spectrophotometer.

Standard solution.—0·2786 g of bismuth oxide (B.P.) was dissolved in minimum volume of concentrated hydrochloric acid. The solution was then boiled to white fumes with 1 ml of concentrated sulphuric acid and a drop or two of concentrated nitric acid. The resulting mass was dissolved in 2 N sulphuric acid and was made upto 250 ml and used as the stock solution (0·1 g/100 ml). A 0·01 M solution of tetraphenylarsonium chloride (Fluka A. G., Buchs SG) and freshly prepared (1·6%) solution of potassium iodide (B.D.H.), both in distilled water, were employed as the reagents.

Procedure.—An aliquot of the standard solution containing 10 μg to 100 μg of bismuth was taken in different separating funnels. The volume in each was made upto 5 ml by adding 2 N sulphuric acid. To each of these solutions 1 ml of KI reagent followed by 1 ml of tetraphenylarsonium chloride reagent was added. The resulting mixture having orange yellow precipitate was then shaken for 1 minute with 3 ml of 1, 2-dichloroethane. The organic layer was separated and the absorbance was measured at 492 nm against the pure solvent. Alternatively, the orange coloured precipitate formed, after the addition of the reagent was centrifuged and the supernatant liquid was drained off and the precipitate was then dissolved in 3 ml of 1, 2-dichloroethane and its absorbance measured at 492 nm. The orange coloured extract was found to obey Beer's law over the concentration range of 3 to 30 μg/ml. The experiments were repeated with varying amounts of bismuth and the amounts of bismuth were calculated from the standard calibration curve. The results of these determinations showed that the error involved was less than 3%.

In a similar manner, 50 μg of bismuth was estimated in presence of the metal ions Fe^{3+}, Cd^{2+}, Sn^{4+}, Hg^{2+} and Sb^{3+} and the anions $AuCl_4^-$, MoO_4^{2-}, WO_4^{2-}, CrO_4^{2-}, MnO_4^{4-}, CNS^-; $S_2O_8^{2-}$, F^-, $C_2O_4^{2-}$ and ClO_4^- in amounts equal to and 10 times that of the bismuth added. The recovery of bismuth, in most cases, in presence of equal amounts of interfering ions was good. A 10 fold excess of some of the ions interfered and a few interfered even when present in equal amount.

Discussion

Bismuth, in presence of potassium iodide, forms orange yellow precipitate of tetrapheylarsonium tetraiodobismuthate with tetraphenylarsonium chloride extractable into 1, 2-dichloroethane. The orange coloured extract has an absorbance maximum at 492 nm and obeys Beer's law over the concentration range 3 to 30 μg per ml. The maximum error involved in the determination of trace quantities of bismuth in absence of other ions is within 3%.

An examination of the effect of the presence of other metal ions, *viz.*, Fe(III), Cd(II), Sn(IV), Hg(II) and Sb(III) that form halide complexes showed that these metals when present in equal amounts with bismuth did not interfere much (error within 5% in the μg level). However, except Hg(II) and Sn(IV), all the other three in their 10 fold excess interfered. A similar study on the effect of the presence of common anions, *viz.*, $AuCl_4^-$, MoO_4^{2-}, WO_4^{2-}, CrO_4^{2-}; MnO_4^- CNS^-,

$S_2O_8^{2-}$, F^-, $C_2O_4^{2-}$ and ClO_4^- showed that only CrO_4^{2-} and MnO_4^- interfered seriously when present in equal amounts and in 10 fold excess to that of the bismuth. $S_2O_8^{2-}$, $AuCl_4^-$ and CNS^- ions in 10 fold excess interfered, the latter two interfered only slightly. All other anions did not interfere even when present in 10 fold excess.

Interference due to iron (III) could be removed by using oxalic acid and ammonium fluoride as the masking reagents and a good recovery of bismuth (error within 1%) was achieved. These reagents did not interfere in the determination of bismuth. Interference due to chromium (VI) could be removed by prior separation of chromium by extraction with isobutyl methyl ketone from solutions above 1 N with respect to hydrochloric or sulphuric acid, a method that has been used in the determination of chromium[11,12]. Recovery of bismuth was within 2% error. The application of the common analytical practice of reducing potassium permanganate with oxalic acid in acidic medium proved to be successful in the removal of interference due to MnO_4^- ion. Recovery of bismuth was within 3% error.

Thus the method proved to be a simple and rapid one providing a good recovery of bismuth in presence of equal amounts of most of the common ions. Besides, it also provides simple techniques for the removal of interferences due to these interfering seriously.

The authors express their sincere thanks to Prof. N. N. Siddhanta, for providing laboratory facilities in carrying out this work.

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1. Willard, H. H. and Smith, C. M., *Ind. and Eng. Chem. Anal. Ed.*, 1939, 11 (4), 186.
2. —, *Ibid.*, 1939, 11 (5), 269.
3. —, *Ibid.*, 1939, 11 (6), 305.
4. Tribalat, S., *Anal. Chim. Acta,* 1949, 3, 113 ; 1950, 4, 288.
5. Neeb, R., *Z. Anal. Chem.*. 1957, **154**, 17.
6. Affsprung. H. E. and Robinson, J. L., *Anal. Chim. Acta,* 1967, 37 (1), 81.
7. Robinson, J. L., *Univ. Microfilms (Ann. Arbor, Mich)*, Order No. 66–14, 244, 147 ; *Diss. Abstr.*, 1966, B 27 (6), 1762.
8. Magee, R. J. and Khattak, M. A., *Microchim. J.*, 1964, 8 (3), 285.
9. Murphy, J. W. and Affsprung, H. E., *Anal. Chim.*, 1961, 33, 1658.
10. Nobeohisa, M. and Akira, K., *Trans. Natt. Research,* 1959, 1, 69.
11. Hedley, A., *Analyst,* 1968, 93, 289.
12. Baishya, N. K. and Heslop, R. B., *Anal. Chim. Acta,* 1970, 51, 69.

MESOMORPHIC BEHAVIOUR OF SOME SCHIFF BASE COMPOUNDS COMPRISING NAPHTHALENE MOIETY

A number of homologous series comprising schiff base compounds exhibiting mesomorphism are known. Generally a linear structure favours the development of mesomorphism. It is observed that when molecular structure is varied within a group of suitable compounds the liquid crystalline properties change markedly. The introduction of a 1, 4 substituted naphthalene nucleus in place of a 1, 4 substituted phenylene ring usually reduces the thermal stabilities of the mesophase.

Dave *et al.*[1-5], have studied a number of schiff base series comprising a naphthalene moiety. As a part of our programme of the study of the effect of breadth on mesomorphism we describe here the mesomorphic behaviour of the four compounds of the series 4-*n*-alkoxy-1-naphthalidene-4′-aminobiphenyls. The melting points and transition temperatures are summarized in Table I.

Table I

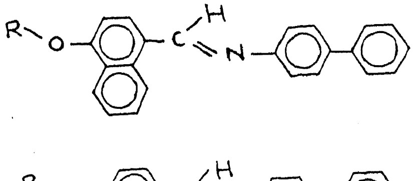
4-n-Alkoxy-1-naphthylidene-4′-aminobiphenyls

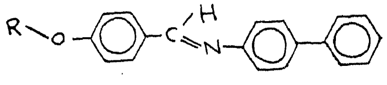
$RO.C_{10}H_6CH : N.C_6H_4.C_6H_5$

R	Transitions temperatures (° C)		
	Smectic	Nematic	Isotropic
1. CH_3	..	..	201·5
2. $C_{12}H_{25}$	..	(103·0)	107·5
3. $C_{16}H_{33}$	..	92·0	97·0
4. $C_{18}H_{37}$	(69·0)	83·5	93·0

Values in parenthesis indicate monotropy.

The methyl derivative is non-mesomorphic ; the dodecyl derivative exhibits a monotropic nematic mesophase and the last two members, *i.e.*, hexadecyl and octadecyl derivatives exhibit enantiotropic nematic mesophases. The octadecyl derivative exhibits a monotropic smectic mesophase also.


A


B

The nematic mesophase exhibited by these compounds has the threaded texture and the smectic mesophase has the broken fan shaped texture, The mesomorphic thermal stabilities of the compounds of the present series (series A) are comparatively less than those of the corresponding compounds of the benzylidene series, *p*-*n*-alkoxybenzylidene-4-aminobiphenyls[6] (series B). This is expected as series (A) contains a 1, 4 substituted naphthalene nucleus at one end of the molecule which increases the molecular breadth resulting in the decrease of the thermal stabilities of the mesophases. It is of interest to note that only the last member of the series (A), *i.e.*, the octadecyl derivative exhibits smectic mesophase which is monotropic in nature. In the case of series (B) the smectic phase appears at the pentyl derivative and purely smectic in nature. The reason for this late from the dodecyl derivative onwards the series is commencement of smectic mesophase in series (A) can be attributed to the increased breadth of the molecules of series (A) due to the introduction of the naphthalene moiety which not only reduces the thermal stabilities of the mesophases but also delays the commencement of the smectic mesophase.

The authors take this opportunity to express their thanks to Professor S. M. Sethna, for his keen interest in the work.

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1. Dave, J. S., Kurian, G., Prajapati; A. P. and Vora, R. A., *Mol. Cryst. Liquid Cryst.*, 1971, 14, 307.
2. —, —, — and —, *Indian J. Chem.*, 1972, 10, 754.
3. —, —, — and —, *Curr. Sci.*, 1972, 41, 415.
4. —, and Prajapati, A. P., Paper presented at the International Liquid Crystal Conference, Bangalore, 1973; (in press, *Pramāṇa* Bangalore, India).
5. — and —, unpublished work.
6. Gray, G. W., Hartley, J. B., Ibbotson, A. and Jones, B., *J. Chem. Soc.*, 1955, p. 4359.

ION-EXCHANGE KINETICS STUDY USING HYDROUS CERIC OXIDE AS AN ION-EXCHANGER

STUDY of the kinetics of ion-exchange is of importance in the use of ion-exchange resins for chemical separations. There is, however, a paucity of such data on inorganic ion-exchangers[1]. The present paper describes an investigation of the kinetics of exchange reactions in hydrous CeO_2 using ammines of Co^{3+}, Ni^{2+}, Cu^{2+}, Zn^{2+} and Cd^{2} as exchanging ions. The study includes (1) dependence of the exchange rate on the concentration of the exchanging ion and (2) effect of the size of the exchanger particle and exchanging ion on the rate of exchange.

Materials and Method

All the reagents used were of A.R. quality. Hydrous CeO_2 was prepared by adding excess of aqueous ammonia to a saturated solution of ceric ammonium sulphate[2]. The resulting precipitate was washed free of sulphate and dried at 100–110° C. Ammine complexes of Co^{3+}, Cu^{2+} and Ni^{2+} were prepared by the usual methods[3] and those of Zn^{2+} and Cd^{2+} by dissolving appropriate quantities of the metal salts in water followed by the addition of excess of aqueous ammonia.

Procedure for the Rate-Study.—Samples of 20 ml of the metal ammines were added to a series of stoppered conical flasks, each containing 100 mg of the exchanger. The resulting mixtures were shaken mechanically up to equilibrium. The solutions were immediately centrifuged and the supernatants (5 ml) were analysed for Co^{3+} and Cu^{2+} iodometrically[4,5] and Ni^{2+}, Cd^{2+} and Zn^{2+}, complexometrically[6] and the amount of metal ions sorbed, hence their F-values were found out from the relation[7]

$$F = \frac{\text{Amount of the metal ion sorbed at time } t}{\text{Total amount of the metal ion sorbed at equilibrium}}$$

The samples of CeO_2 with an average particle radius $9 \cdot 0 \times 10^{-3}$ cm and $0 \cdot 85 \times 10^{-3}$ cm were obtained with the help of standard sieves. The particle sizes were further checked by microscopic measurements.

Results and Discussion

The results presented in Tables I and II indicate that

(*i*) The ion-exchange kinetics exhibit two distinctly separate stages, relatively quick process in the first stage followed by a slower one towards the end.

(*ii*) Smaller the size of the exchanger particle as well as exchanging ion faster is the rate of exchange.

(*iii*) Amongst the ammine complexes under study, the exchange rate follows the order :

$$Co > Cu > Zn > Ni > Cd.$$

It is well known[8] that sorption of metal ions on hydrous oxides takes place by the mechanism of ion-exchange. It is also known[9] that the overall exchange process is mainly governed by two types of diffusion processes. The first process, *i.e.*, film diffusion depends on (*a*) stirring characteristics, (*b*) the external concentration of the exchanging ions and (*c*) the size of the exchanger particle. The second process, *i.e.*, particle diffusion depends only on the size of the exchanger particle[10]. In very dilute solutions the rate of diffusion of electrolyte across the film barrier about the

TABLE I

Dependence of F-values on the concentration of different ammines

Particle radius = 9·0 × 10^{-3} cm. Amount of the exchanger = 100 mg.
Total volume = 20 ml, pH = 10·5

Time	Fractional attainment of equilibrium (F)									
	$Co(NH_3)_6^{3+}$		$Cu(NH_3)_4^{2+}$		$Zn(NH_3)_4^{2+}$		$Ni(NH_3)_6^{2+}$		$Cd(NH_3)_4^{2+}$	
	0·05 M	0·10 M	0·05 M	0·10 M	0·05 M	0·10 M	0·05 M	0·10 M	0·05 M	0·10 M
30 secs	0·09	0·20	0·015	0·11	..	0·10	..	0·05	..	0·02
1 min	0·13	0·36	0·03	0·14	0·02	0·13	0·01	0·12	0·025	0·10
2 min	0·20	0·42	0·09	0·40	0·08	0·39	0·05	0·30	0·075	0·25
5 min	0·34	0·60	0·25	0·55	0·28	0·54	0·26	0·52	0·21	0·50
10 min	0·60	0·70	0·36	0·66	0·45	0·65	0·42	0·64	0·40	0·62
20 min	0·70	0·80	0·62	0·80	0·70	0·78	0·65	0·76	0·625	0·75
30 min	0·82	0·86	0·74	0·85	0·81	0·84	0·78	0·82	0·765	0·81
45 min	0·89	0·89	0·82	0·88	0·89	0·89	0·88	0·87	0·86	0·86
60 min	0·93	0·93	0·89	0·92	0·925	0·925	0·91	0·91	0·90	0·90
75 min	0·95	0·95	0·93	0·94	0·95	0·95	0·94	0·94	0·93	0·93
90 min	0·97	0·97	0·96	0·96	0·97	0·97	0·96	0·96	0·96	0·96
100 min	0·98	0·98	0·98	0·98	0·98	0·98	0·98	0·98	0·98	0·98
110 min	1·00	1·00	1·00	1·00	0·99	0·99	0·99	0·99	0·99	0·99
120 min	1·00	1·00	1·00	1·00	1·00	1·00	1·00	1·00	1·00	1·00

TABLE II

Dependence of F-values on the concentration of different ammines

Particle radius = 0·85 × 10^{-3} cm. Amount of the exchanger = 100 mg. Total volume = 20 ml, pH = 10·5

Time	Fractional attainment of equilibrium (F)									
	$Co(NH_3)_6^{3+}$		$Cu(NH_3)_4^{2+}$		$Zn(NH_3)_4^{2+}$		$Ni(NH_3)_6^{2+}$		$Cd(NH_3)_4^{2+}$	
	0·05 M	0·10 M	0·05M	0·10 M	0·05 M	0·10 M	0·05 M	0·10 M	0·05 M	0·10M
30 secs	0·31	0·32	0·18	0·24	0·10	0·18	0·015	0·14	0·01	0·05
1 min	0·41	0·40	0·20	0·38	0·12	0·36	0·025	0·28	0·04	0·10
2 min	0·45	0·46	0·28	0·49	0·15	0·47	0·08	0·39	0·10	0·35
5 min	0·66	0·65	0·42	0·57	0·27	0·55	0·24	0·53	0·26	0·50
10 min	0·80	0·82	0·58	0·74	0·47	0·72	0·48	0·70	0·52	0·68
20 min	0·92	0·94	0·79	0·90	0·72	0·88	0·71	0·85	0·76	0·83
30 min	0·99	0·99	0·90	0·93	0·86	0·94	0·84	0·92	0·87	0·90
45 min	1·00	1·00	0·96	0·99	0·96	0·98	0·96	0·965	0·95	0·95
60 min	1·00	1·00	0·99	1·00	1·00	1·00	0·99	1·00	0·98	0·98
75 min	1·00	1·00	1·00	1·00	1·00	1·00	1·00	1·00	0·99	0·99
90 min	1·00	1·00	1·00	1·00	1·00	1·00	1·00	1·00	1·00	1·00
100 min	1·00	1·00	1·00	1·00	1·00	1·00	1·00	1·00	1·00	1·00
110 min	1·00	1·00	1·00	1·00	1·00	1·00	1·00	1·00	1·00	1·00
120 min	1·00	1·00	1·00	1·00	1·00	1·00	1·00	1·00	1·00	·001

exchanger is rate-controlling. As the concentration is increased, the kinetics is determined by both the diffusion across the film and the diffusion in the solid. At still higher concentrations, particle diffusion becomes the sole rate determining step[9] and the rate of diffusion becomes concentration independent[11], *i.e.*, F becomes independent of concentration. Our results (Tables I and II) show both film and particle diffusion are responsible for the overall exchange process. Since, F-values initially increase with the increase of exchanging ion concentration from 0·05 M to 0·10 M, followed by a more or less identical F-values.

From Tables I and II it is seen that the exchange rate in smaller (0·85 × 10^{-3} cm) particle is very much faster than the same in the bigger (9·0 × 10^{-3} cm) size particles due to the increase in specific surface area.

The faster rate of exchange of Co (III)-ammine as compared to the other metal ammines under study may be explained on the basis of their known covalent radii[12,13,14] ; Octahedral Co (III), 1·22 Å and Ni (II), 1·39 Å; Tetrahedral Zn (II), 1·31 Å and Cd (II), 1·48 Å; square planer Cu (II), 1·28 Å. Therefore their sizes follow the order:

$$Co(NH_3)_6^{3+} < Cu(NH_3)_4^{2+} < Zn(NH_3)_4^{2+} < Ni(NH_3)_5^{2+} < Cd(NH_3)_4^{2+}$$

From our results (Tables I and II) it seems smaller the size of the exchanging ion, faster will be the rate of exchange through the pores of the exchanger solid, resulting in a faster rate of exchange.

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Department of Chemistry, A. K. BHADURI.
University of Delhi, K. R. KAR.
Delhi 110 007, *August* 18, 1975. K. B. PANDEYA.

1. Marinsky, J. A. and Marcus, Y., *Ion Exchange and Solvent Extraction,* Marcell Dekker, Inc., New York, Vol. V, 1973, p. 122.
2. Cotton, F. A. and Wilkinson, G., *Advanced Inorganic Chemistry,* 2nd ed., Wiley Eastern Pvt. Ltd., 1970, p. 1067.
3. Marr, G. and Rockett, B. W., *Practical Inorganic Chemistry,* Reinhold, New York, pp. 266, 270, 277.
4. — and —, *Ibid.,* p. 267.
5. Glethorpe, O. and Smith, *Analyst,* 1943, 68, 325.
6. Flaschka, H. A., *Titrations. EDTA,* Pergamon Press, Oxford, 1964, pp. 80, 83.
7. Kunin, R., *Elements of Ion-Exchange,* Reinhold, 1960, p. 12.
8. Vesely, V. and Pekarek, V., *Talanta,* Pergamon Press, Northern Ireland, Vol. 19, 1972, p. 220.
9. Kunin, R., *Elements of Ion-Exchange,* Reinhold, 1960, p. 16.
10. Conway, D. E., Green, J. H. S. and Reichenberg, D., *Trans. Faraday Soc.,* 1954, 50, 511.
11. Amphlett, C. B., *Inorganic Ion-exchangers,* Elsevier, New York, 1964, p. 128.
12. Pauling, L., *The Nature of the Chemical Bond,* Third ed., Cornell University Press, New York, 1963, pp. 249, 252.
13. Heslop, R. B. and Robinson, P. L., *Inorganic Chemistry,* Third ed., Elsevier, New York, 1967, p. 743.
14. — and —, *Ibid.,* 1967, pp. 702, 705, 741, 756.

ADSORPTION OF HYDROGEN ON PRECIPITATED FISCHER-TROPSCH IRON CATALYSTS

ADSORPTION of hydrogen has been studied on two catalysts containing originally (A) Fe_2O_3, CuO and K_2O in the ratio of approximately 90 : 8 : 0·4 and (B) Fe_2O_3 and K_2O in the proportion of 98 : 0·4 respectively. These catalysts are being tried in the Fischer-Tropsch synthesis of hydrocarbons at the Institute.

The catalysts were prepared by precipitation with Na_2CO_3 from a solution of the nitrates of the metals. The dry precipitate was reduced at 300° C for 6 hr.

The adsorption of hydrogen was measured in a volumetric gas adsorption unit in the temperature range 99°–275° C. The time for equilibration at each pressure was one hour. The degassing of the catalysts was carried out at 400° C, for 5 hr.

The adsorption isotherms in the case of both catalysts exhibit, as shown in Figs. 1 and 2, an increase of adsorption with increase in temperature upto about 210° C, and then a decrease. The surface area of the catalysts, as measured by the adsorption of argon at liquid nitrogen temperature, using the approach of de Boer and coworkers[1], was

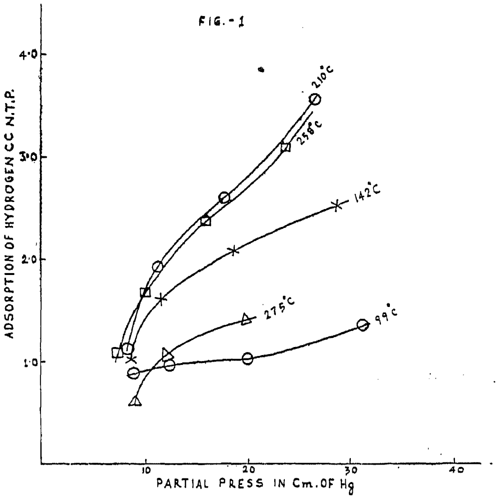

FIG. 1. Adsorption isotherms of hydrogen on catalyst (A) (7·5 g. dry).

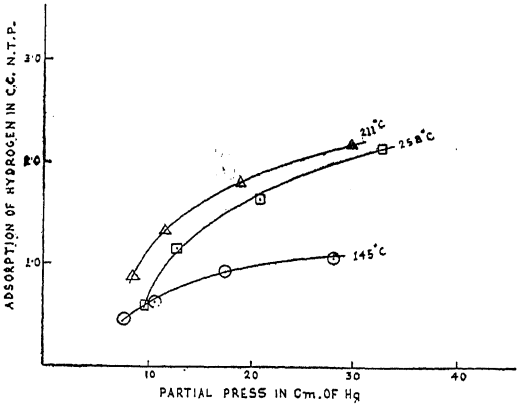

FIG. 2. Adsorption isotherms of hydrogen on catalyst (B) (6·65 g. dry).

7·5 and 8·0 sq. meters/g respectively for catalysts, (A) and (B). The amounts of hydrogen adsorbed, however, were lower with catalyst (B) than with (A). The surface coverage at maximum of adsorption on catalyst (A) was approximately 80%, assuming an atomic radius of hydrogen of 2·08 Å. It appears from the result that the role of copper in the catalyst lies in increasing the adsorption of hydrogen.

Heats of adsorption of hydrogen were determined isosterically on catalyst (A) in the region 210–275° C. The values increased from 3·5 to 3·9 k.cal/mole for coverages of 0·07 to 0·12 cm^3/g. These results compare favourably with a value of 3·9 k.cal/mole calculated theoretically using the CFSO–BEBO model[2,4].

The kinetics of adsorption on catalyst (A) at 142° C and 210° C obey the Elovich[3] equation. The energy of activation of hydrogen adsorption calculated from the rate data at 142° C and 210° C, at 1·2 c.c. N.T.P. of adsorption, was 1·4 k.cal/mole, showing that we are dealing with a chemisorption of low energy of activation.

Central Fuel Research Institute, B. K. SHARMA.
Dhanbad, Bihar, *August* 28, 1975. K. A. KINI.

---

1. Lippens, B. C., Linsen, B. G. and DeBoer, J. H., *Journal of Catalysis*, 1964, 3, 32.
2. Weinberg, W. G. and Merill, R. P., *Surface Science*, 1972, 33, 493.
3. Elovich, S. Y. and Zhabrova, G. M., *Zhur. fiz. khim.*, 1939, 13, 1716, 1775.
4. Kini, K. A., *Journal of the Indian Chemical Society*, 1975, 50, 1106.

EXTRACTION AND PHOTOMETRIC STUDIES OF Cu (II) WITH N-BENZOYL-N′-METHYL THIOUREA

THE complexation reaction of Cu(II) with substituted thioureas is known. Banerji and Sukhatankar[1] have reported complex compounds of mono and di-N-phenyl thiourea with some copper salts. The present work reports the extraction and photometric studies of copper(II) with N-benzoyl-N′-methyl thiourea (abbreviated as BMT). BMT required in the present work was prepared by the action of benzoyl isothiocyanate with methylamine in acetone medium[2,3]. It was crystallized from alcohol water mixture and the purity was confirmed by the composition studies and IR spectra. AnalaR grade copper sulphate was used to prepare Cu (II) solution.

General procedure for extraction.—The pH of a solution containing 1000 µg of Cu (II) was adjusted to 6·0 in a total volume of 25 ml. This was shaken for two minutes each with two, 5 ml portions of BMT in benzene (2000 µg/5 ml). The yellowish green coloured complex in the two benzene extracts was mixed and the total volume was made upto 10 ml with benzene. The absorbance of this solution was then measured at 355 nm against reagent blank.

Results and Discussion :

The complex shows λ_{max} at 355 nm against reagent blank. The absorbance measurements were made at this wavelength. The system conforms to Beer's law in the concentration range of 1 to 5 mg of copper. The complex remains unchanged for two hours as indicated by the absorbance.

The extraction was carried out at different pH values. Quantitative extraction takes place at pH 6·0. There is no extraction below pH 4·5 while above pH 6·4, copper gets hydrolysed. Various solvents were tried for extraction but benzene was found to be the best.

The effect of varying reagent concentration on the extraction was studied. The absorbance increases upto the ratio 1 : 4 (Cu : BMT), and then becomes practically constant. This shows that four fold excess of the reagent is necessary for the quantitative extraction. The presence of alkali nitrates and chlorides did not enhance the extraction.

The ratio of Cu (II) : BMT in the extracted species was determined by the method of continuous variations modified for a two phase system by Irving and Pierce[4]. The composition was 1 : 2 [Cu(II) : BMT]. This was further confirmed by mole ratio method.

The effect of various ions on the extraction was also studied. The tolerance limit for each ion was calculated at that concentration where a change of ± 2% in the extraction was noted. Table I shows the results obtained.

TABLE I
Effect of diverse ions

Tolerance limit	Ion present
$1 \cdot 0 \times 10^3$	Na^+, Ba^{2+}, Br^-, $Cr_2O_7^{2-}$
$5 \cdot 0 \times 10^2$	Mg^{2+}, Ca^{2+}, Cr^{6+}, UO_2^{2+}, K^+, PO_4^{3-}, $S_2O_3^{2-}$, SCN^-, EDTA, NO_2^-, Cl^-, I^-, tartarate
$1 \cdot 0 \times 10^2$	Mn^{2+}, Zn^{2+}, Pd^{2+}, Cd^{2+}, Pb^{2+}, Bi^{3+}, Th^{4+}, $C_2O_4^{2-}$, citrate, CH_3COO^-, ClO_4^-, malonate.

V^{5+}, Fe^{3+}, Co^{2+}, Ni^{2+} and Zr^{4+} interfere at all concentrations.

From ten determinations, the absorbance comes to be 0·26 ± 0·01. The relative mean deviation is 2·68%.

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Nagpur 440 010, *August* 12, 1975.

---

1. Banerji, S. N. and Sukthankar, S. C., *J. Ind. Chem. Soc.*, 1963, 40 (5), 387.
2. Douglass, I. B. and Dane, F. B., *J. Amer. Chem. Soc.*, 1934, 56, 719.
3. Nair, G. V., *J. Ind. Chem. Soc*, 1963, 40, 953.
4. Irving, H. and Pierce, T. B., *J. Chem. Soc.*, 1959, p. 2565.

A PRELIMINARY REPORT ON THE OCCURRENCE OF RADIOLARIA IN THE MIDDLE EOCENE OF THE VINJHAN-MIANI AREA, KUTCH, GUJARAT

THIS note records for the first time a radiolarian genus *Sethodiscinus* Haeckel, 1887 (Figs 1–3) from the fossiliferous yellow limestone exposed 1·6 km. NEE of the Khirasra village (N.L. 23° 7′ 5″ : E.L. 69° 2′ 15″) in a nala flowing towards the Khirasra village. The planktonic foraminifera are identified as *Globorotalia lehneri*, *Globigerina tripartita topuriensis*, *Globanomalina micra*, *Inordinatosphaera indica*, *Chiloguembelina martini* and *C. tenuis* from the fossiliferous yellow limestone by one of the authors (A. K. J.). On the basis of the occurrence of these planktonic foraminifera this limestone has been referred to Lutetian. Srivastava[1] noted the following geological sequence of beds in the Vinjhan–Miani area :

Lutetian {
Fossiliferous light yellow marl
Fossiliferous yellow limestone
Unfossiliferous khaki shales

Jauhari[2,3], studied the planktonic as well as benthonic foraminifera obtained from the fossiliferous light yellow marl in detail and assigned a Lutetian age to this marl.

Systematic Palaeontology

Superfamily	CENODISCICAE Haeckel, 1887
Subsuperfamily	CENODISCILAE Haeckel, 1887
Family	PHACODISCIDAE Haeckel, 1882
Subfamily	PHACODISCINAE Haeckel, 1882
Genus	*Sethodiscinus* Haeckel, 1887

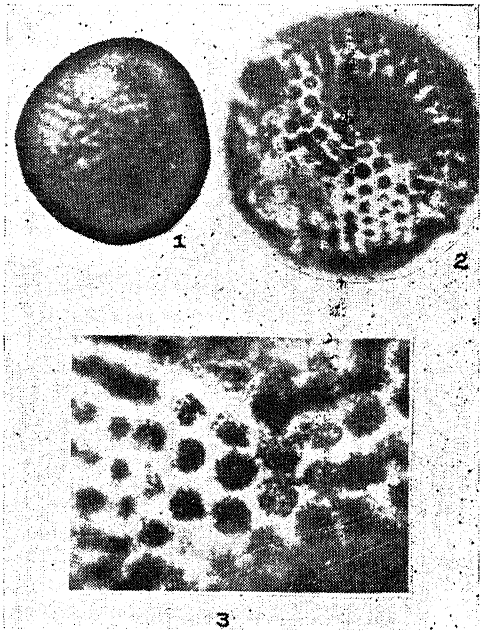
Sethodiscinus sp. indet.
(Figs. 1–3)

Description

Spherical cortical shell with more or less hexagonal net work; medullary shell indistinct. Diameter 0·15 mm.

Remarks

The present form is identical in outline to the genus *Sethodiscinus* Haeckel, 1887 ranging in age from Eocene to Recent. It is rarely distributed. The specimen (Slide No. M.G.D./L.U./K./R./1) has been deposited in the Museum, Department of Geology, University of Lucknow, Lucknow.



FIGS. 1–3. *Sethodiscinus* sp. indet., Fig. 1, surface view of the shell, × 180 ; Fig. 2, surface view of the shell after mounting in natural canada balsam, × 266 ; Fig. 3, enlarged view of the surface of the shell showing more or less hexagonal markings, × 798.

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University of Lucknow, A. K. JAUHARI.
Lucknow, *January* 24, 1975.

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1. Srivastava, I. P., *Unpublished Ph.D. Thesis, Lucknow University*, 1970.
2. Jauhari, A. K., *Curr. Sci.*, 1974, 43 (7), 212.
3. —, *Ibid.*, 1974, 43 (18), 582.
4. Moore, R. C., *Treatise on Invertebrate Paleontology*, Geol. Soc America and Uni, Kansas Press, 1954, Part D, Protista.

ON ACID AND ALKALINE PHOSPHATASE ACTIVITY IN FAT BODIES OF *TENEBRIO MOLITOR* L.[*]

ACID and alkaline phosphatases in the late larval and early pupal fat bodies of *Tenebrio molitor* L. were studied. It is suggested that the phosphatases were concerned mainly with the protein synthesis in fat bodies. Variation in the phosphatase activity of the two stages was recorded.

Observations on studies on the acid and alkaline phosphatase activity in the fat bodies of *Tenebrio molitor* L. are wanting. The present experiments seek variable activity of the two phosphatases in the late larval and early pupal stages of this beetle.

Material and Method

A culture of *T. molitor* was raised on crushed wheat feed at room temperature. Larvae, three days before pupation, and pupae one day after pupation were taken to record acid and alkaline phosphatase activity in the fat bodies. Gomori's techniques (Pearse, 1968), for histochemical demonstration of phosphatases were used. Hartree (1972) was followed for the determination of protein and Tanaka and Iizuka (1968) for acid and alkaline phosphatases. Fat bodies from ten insects were pooled—0·02 ml from each insect sucked up with a micro-syringe—for the determination of acid and alkaline phosphatases and protein.

Results

The acid phosphatase activity in the fat body nucleus was diffused and feeble. The cytoplasm of the fact globule was positive to this phosphatase. The activity near the nucleus was in the form of granules. The cytoplasm away from the nucleus was diffusely positive. The larval fat body exhibited numerous positive granules. After the larval moult, *i.e.,* in early pupal stage the size and intensity of the granules fell significantly. Acid phosphatase activity was $4 \cdot 5 \times 10^{-6}/\mu g$ protein/sec. in larva which reduced to $1 \cdot 7 \times 10^{-6}/\mu g$ protein/sec. in pupa.

The alkaline phosphatase activity in the cytoplasm of the fat mass was also in the form of granules. The nucleus was negative. The nuclear membrane was intensely positive. The outer edge of the cytoplasm was lined with large granules. The early pupal stage showed reduced alkaline phosphatase activity as compared to that of the larval form. Alkaline phosphatase activity in the larva was $6 \cdot 6 \times 10^{-6}/\mu g$ protein/sec. which diminished to $1 \cdot 3 \times 10^{-6}/\mu g$ protein/sec. in pupa.

Discussion

The functional unit of a fat body is composed of a large central fat globule besiezed with a large number of peripheral granules. The latter were called protein storage granules (Locke and Collins, 1965), and acid phosphatase rich granules (Benson and Benson, 1966). Acid phosphatase has been shown to be present in lysosomes. De Duve (1959) considered that the lysosomes are the limiting bags delimited by lipoprotein membranes and they contain acid phosphatase as one of the enzymes. It has been suggested that the acid phosphatase may be concerned with active protein synthesis (Eranko, 1951 ; Pearse, 1968 ; Novikoff, 1961). In the present experiment it was noted that two days before the larva moulted into pupa, the acid phosphatase activity in the fat bodies was very strong and it was reduced noticeably after the moult. Walker (1966) proposed that the release of lysosomal enzymes and therealong reorientation of cellular organelles provide an evidence for the synthesis of proteinaceous food-stuff. Synthesis and release of protein by fat bodies in *Schistocerca gregaria* is shown (Hill, 1965).

It is suggested that alkaline phosphatase activity might be concerned with the nucleic acid synthesis. Staining the fat globules with toluidine blue, Bonhag (1955) suggested that they may contain RNA. Danielli (1953) indicated that the alkaline phosphatase plays an important role in nucleic acid synthesis. It was pertinently noticed in the present experiment that alkaline phosphatase activity was much lowered after the larval moult which might be related with the low cellular metabolic activity. Alkaline phosphatase, probably hydrolyses ATP in fat globule to liberate energy to be used in protein synthesis.

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1. Benson, K. A. and Benson, D. G., *J. Cell. Sc.*, 1966, 1, 81.
2. Bonhag, P. F., *J. Morph.*, 1955, 96, 381.
3. Danielli, J. F., In : *Cytochemistry*, John Wiley and Sons, Publ., Inc., N.Y., 1953.
4. De Duve, C., In : *Subcellular Particles*, Ed., T. Hayashi, 128, Ronald, Pr., N.Y., 1959.
5. Eranko, O., *Acta Physiol. Scand.*, 1951, 24, 1.
6. Hill, L., *J. Insect Physiol.*, 1965, 11, 1605.
7. Hartree, E. F., *Anal. Biochem.*, 1972, 48, 422.
8. Locke, M. and Collins, J. V., *J. Cell Biol.*, 1965, 26, 837.
9. Novikoc, A. B., *The Cell*, Acad. Pr., N.Y., 1961, 2, 423.
10. Pearse, A. G. E., In : *Histochemistry Theor. and Appl.*, 1968.
11. Tanaka, K. and Iizuka, Y., *Biochem. Pharmacol.*, 1968, 17, 2023.
12. Walker, P. A., *J. Insect. Physiol.*, 1966, 12, 1009.

OCCURERNCE OF *NEREIS* (*NEANTHES*) *MOSSAMBICA* IN INDIAN WATERS

STUDIES on the systematics of Polychaetous Annelids of India have been extensive[1-3]. The present finding of a rare Errant Polychaete *Nereis* (*Neanthes*) *mossambica* Day, 1957 (Family: Nereidae) in the brackish waters of Visakhapatnam Harbour is worth reporting. A perusal of the existing literature has revealed that this is the first record of this species from Indian waters. The only one record available to date is the original report by Day[2], from Morrumbene estuary, Mocambique, South Africa. It is hoped that the present report will stimulate search for this Polychaete in other regions of the world in general and India in particular.

The description of the specimens presently collected agrees almost entirely with that given by Day[2] excepting for a few minor variations: Length 45 mm. Width 2 mm. About 75 segments. Proboscis (Figs. 1 and 2) with Group I = 1–3 in a line, Group II = 16 in three oblique rows, Group III = a patch of 21, Group IV = a wedge of 22, Group V = 0, Group VI = one large oval paragnath, Groups VII and VIII = a band of two irregular rows.

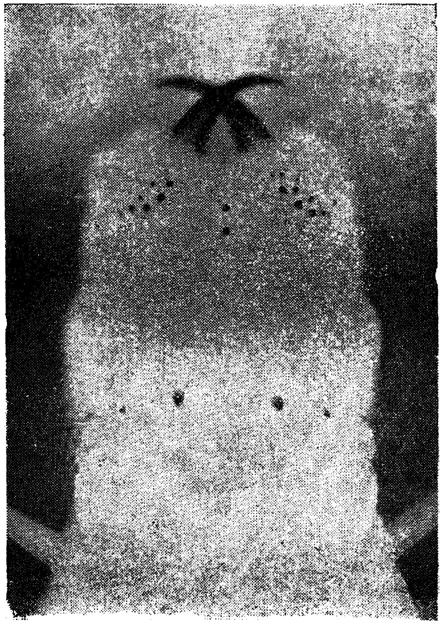

FIG. 1. Proboscis: Dorsal view.

It is possible that the species has a wider distribution than was originally believed and efforts to look for it both along the East and West coasts of the Indian peninsula would be rewarding.

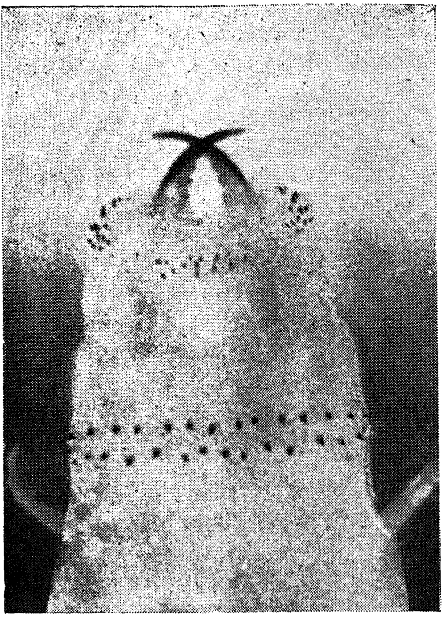

FIG. 2. Proboscis: Ventral view.

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Andhra University, Waltair, *August* 6, 1975.

---

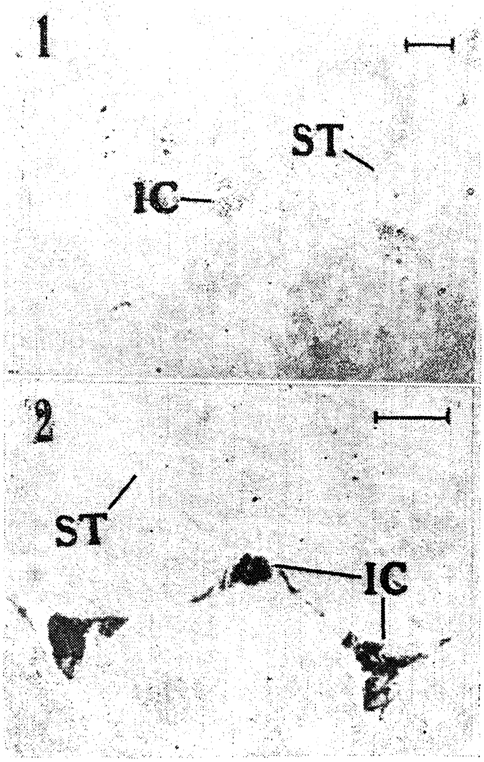
1. Bhatt, Y. M. and Bal, D. V., *J. Univ. Bombay*, 1966, 32 (3 and 5), 1964, 1966, p. 24.
2. Day, J. H., *A Monograph on the Polychaeta of Southern Africa. I. Errantia*, 1967.
3. Parulekar, A. H., *J. Bomb. Nat. Hist. Soc.*, 1972, **68**, 726.

HISTOCHEMICAL STUDY OF THE STEROIDOGENIC SITES IN THE TESTES OF THE LIZARDS, *HEMIDACTYLUS FLAVIVIRIDIS* AND *MABUYA CARINATA*

HISTOCHEMICAL demonstration of Δ^5-3 β-hydroxysteroid dehydrogenase (Δ^5-3 β-HSDH) has shown that the Leydig cells are the active sites of steroidogenesis in the testes of a few reptiles that have been studied so far[1-3]. Recently, besides Δ^5-3 β-HSDH, another key enzyme, 17 β-HSDH involved in the bioconversion of sex steroids, has been demonstrated in the Leydig cells and the seminiferous epithelium in the testes of chameleon, python, crocodile[4] and cobra[5]. However, according to an earlier report, no Δ^5-3 β-HSDH activity could be histochemically observed in the testis of *Hemidactylus flaviviridis*[6]. From the presence of sudanophilic and cholesterol positive lipids, it has been suggested that the Leydig cells of *Mabuya macularica* are the steroid secretors[7]. In view of these observations, histochemical localization of Δ^5-3 β-HSDH, 17 β-HSDH, glucose-6-phosphate dehydrogenase (G-6-PDH) and NADH diaphorase activity was carried to investigate precisely the steroidogenic cellular sites in the testes of the lizards, *H. flaviviridis* and *Mabuya carinata.*

Fresh frozen sections of *H. flaviviridis* and *M. carinata* testes were incubated in the media containing the individual substrates, pregnenolone and dehydroepiandrosterone for localising Δ^5–3β–HSDH : testosterone and 17 β-estradiol for 17 β-HSDH ; D–glucose-6-phosphate disodium salt for G–6–PDH ; and reduced NAD for NADH diaphorase, according to the method described earlier[4,8].

Histochemical reaction for these enzymes in the testes of the two lizards, as indicated by the deposition of dark blue diformazan granules, was intense in the Leydig cells (Figs. 1, 2). The seminiferous epithelium also showed faint but positive reaction for these enzymes. Our observation, the presence of Δ^5-3β-HSDH activity in the testis of *H. flaviviridis,* is contrary to the earlier report that this enzyme is absent in the testis of *H. flaviviridis.* Furthermore, it is interesting to note that the seminiferous epithelium is also capable of steroid biosynthesis as inferred from earlier histochemical observations on a few reptiles[4,5] and the *in vitro* conversion studies on the isolated seminiferous tubules of the cobra testis[9].



FIGS. 1–2. Fig. 1. Showing Δ^5-3 β-HSDH activity in the testis of *H. flaviviridis.* Fig. 2. Showing G–6–PDH activity in the testis of *M. carinata.* (IC = Leydig cells ; ST = Seminiferous tubule. The scale lines indicate 100 μ.)

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Dharwar 580 003, India, *August* 12, 1975.

1. Nandi, J., *Amer. Zool.,* 1967, **7**, 115.
2. Erpino, M. J., *Gen. Comp. Endocr.,* 1971, **17**, 563.
3. Chieffi, G., *Amer. Zool.,* 1972, **12**, 207.
4. Gouder, B. Y. M. and Nadkarni, V. B., *Indian J. Exp. Biol.,* 1974, **12**, 326.
5. — and —, *Curr. Sci.,* 1974, **43**, 785.
6. Prasad, M. R. N. and Sanyal, M. K., *Second Int. Congr. Horm. Steroids, Excerpta Medica Int. Congr.,* 1966, **132**, 1058.
7. Upadhyay, S. N. and Guraya, S., *Gen. Comp. Endocr.,* 1972, **19**, 88.
8. Baillie, A. H., Ferguson. M. M. and Hart, D. Mck, In : *Developments in Steroid Histochemistry,* Academic Press, London, 1966.
9. Lofts, B. and Bern, H. A., In : *Steroids in Nonmammalian Vertebrates,* Ed. by Idler, D. R., Academic Press, New York, 1972.

ANNUAL MALE REPRODUCTIVE CYCLE OF HOUSE SPARROW *PASSER DOMESTICUS* (LINNAEUS)

SEASONAL reproductive cycle of birds has been studied by a number of workers[1,2]. However, such studies on avian species inhabiting the North Western part of the semiarid regions of the Indian subcontinent have not been made. This report embodies observations on the monthly changes in the weight and histology of the adult testes of house sparrows. Monthly collections of the sexually adult males were made. After obtaining the body weight, both the testes were removed and weighed together to the nearest miligram. Portions of the testicular tissues were fixed in Bouin's fluid, embedded in paraffin, sectioned at 5 μ, and stained with Harris haematoxylin and eosin. Tubule diameters were also recorded for each month from the histological sections passing through the center of the testes. At least 6 birds were sacrificed every month.

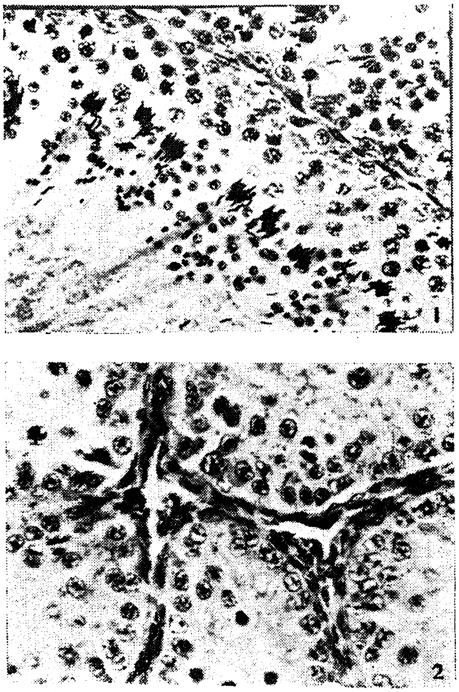
TABLE I

Monthly variations in the average mean body weight, the combined testicular weight, mg testis weight/gm body weight and the seminiferous tubule diameter of the house sparrow Passer domesticus

Month	No. of birds	Body weight (gm)	Testes weight (mg)	mg Testes wt/gm body weight	Tubule Diameter (μ)
January	6	18·84±1·1	35·2± 3·6	1·96± ·022	1·1
February	6	20·16± ·01	215·6± 5·5	10·65± ·092	2·3
March	6	21·1 ±0·8	385·4±31·2	18·26±1·68	3·2
April	6	21·84±0·6	408·2±30·4	19·73±1·46	3·1
May	6	22·28±0·9	441·6±38·1	20·05±2·11	3·2
June	6	20·76±0·6	226·0± 3·9	10·62±0·61	2·1
July	6	21·26±1·4	189·2±17·4	9·12±0·30	1·3
August	6	18·84±1·3	56·4± 6·8	2·99±0·08	1·2
September	6	19·9 ±0·9	14·3± 2·9	0·57±0·013	1·9
October	6	18·6 ±0·8	15·5± 3·2	0·90±0·02	1·0
November	6	20·1 ±0·8	8·8± 1·1	0·44±0·01	0·4
December	6	20·0 ±0·4	5·0± 0·02	0·29±0·001	0·4

Table I presents the monthly variations in the body weight, the testicular weight, mg testis weight/gm body weight and the seminiferous tubule diameter of house sparrows. The testes are sexually regressed and exhibit minimum weight from September to December. The testicular weight begins to increase from January. From February onwards the testicular weight increase rapidly and reaches the maximum in May. The peak in the testicular weight is followed by a sudden, statistically significant ($P < 0{\cdot}001$), decline in June; the combined testicular weight drops from 441·6 mg in May to 226·0 mg in June. This decrease in the testicular weight continues upto December when a fresh cycle starts.

Testicular histology compliments with the weight changes. In January, the majority of birds are sexually inactive but some exhibit early stages of recrudescence. In these, the germinal epithelium becomes two or three layers thick and is composed mainly of spermatogonia and the primary spermatocytes. Tunica albuginea is very thick. In February, the seminiferous tubule diameter increases. The testes become spermatogenetically recrudescent containing numerous spermatids. During March, April and May, house sparrow attains full breeding condition. The seminiferous tubule diameter enlarges maximally and shows bundles of spermatozoa with several layers thick germinal epithelium (Fig. 1). Interstitial cells appear to be scanty in number and the tunica albuginea become thin. In June, the testicular regression commences. There is a decrease in the seminiferous tubule diameter. Regression continues from July to August. During this period, all the tubules of a testes do not appear



FIGS. 1–2. Fig. 1. Photomicrograph of an active testis of *Passer domesticus* showing germ cells at various stages of development. Haematoxylin-eosin, × 10 × 40. Fig. 2. Photomicrograph of an inactive testis of *Passer domesticus* showing only spermatogonia and spermatocytes, × 10 × 70.

in the same stage of regression and some even show a few spermatozoa. By September and October the sexual regression is complete. The tunica albuginea shows slight increase in thickness and the intertubular spaces become filled with interstitial cells. Spermatozoa are completely absent. In November and December, the tubule diameter further shrinks in size and the number of interstitial cells appear greatly increased (Fig. 2).

In the present study, it is observed that there is a correlation between the body weight and the weight of gonads, except during the months of November and December (Table I). During these months, the body weight does not correspond with the testicular weight. The testes are dormant whereas the body weight is comparatively higher. This is in agreement with the previously reported findings of Nice[3,4] who has shown that some species gain weight during winter and therefore, the body weight and the testicular weight ratio is negated. The present result also shows a positive correlation between the body weight and tubule diameter. The tubules are maximally expanded in the month of May when the testicular weight is high. A similar relationship has also been reported in spotted Munia[5].

We wish to record our thanks to Prof. A. S. Kapoor for providing various facilities in the department and are grateful to C.S.I.R. (India) for the award of Junior Fellowship to one of us (M. S.).

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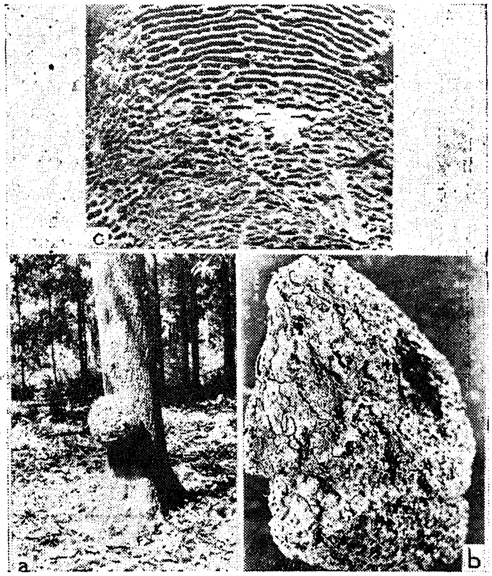
---

1. Loft, B. and Murten, R. K., *J. Zool.*, 1968, 155, 327.
2. Marshall, A. J., *Mem. Soc. Endocr.*, 1955, 4, 75.
3. Nice, M. M., *Trans. Linn. Soc.*, N.Y., 1937, 4, 6.
4. —, *Condor*, 1946, 48, 41.
5. Pandha, S. K., Thapliyal, S. P. and Garg, R. K., *Ind. J. Zool.*, 1973, 1, 117.

PROTECTIVE RESEMBLANCE OF THE NEST OF *NASUTITERMES BECKERI* PRASAD AND SEN-SARMA (INSECTA : ISOPTERA : TERMITIDAE) TO THE BARK OF *VETERIA INDICA**

SEVERAL cases have been cited in literature to illustrate the close resemblance of colour and form of an animal to an object that hormonise with the environment. However, protective resemblance of the nest to an object does not seem to have been reported so far. While surveying the termite fauna of Karnataka State (December 1970), an interesting instance of protective resemblance of termite nest to the bark of *Vateria indica* was observed at Kenchanakumari Reserve Forest. The nests were lodged on the main stem of the trees at heights varying from 50–450 cm. Only one nest was found on one tree. Outer covering of each nest comprised of a thin papery layer exactly matching the colour of the bark of the stem. Colour mimicry of nests was so perfect as to appear as an outgrowth of the tree (Fig. *a*). Each nest was loosely attached to tree and could be taken out intact with little effort. They were light in weight (200 gm for 5650 cc in volume).



FIGS. *a–c*

Cross-section of nests revealed a compact mass with a large number of inter-connected cells or cavities arranged concentrically (Figs. *b, c*). Measurements of individual cells varied from 1·3 to 8·60 × 2·32 cm. The wall separating one cell from the other was very thin (0·03 to 0·05 cm). In addition to the vertical partition walls, each cell had pillars and transverse walls of the same thickness of the partition wall of the cell. These add to the rigidity of the cell. Towards the periphery of the nest, the cells were narrower than in the centre.

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VIABILITY OF *PLASMOPARA VITICOLA* SPORANGIA PRODUCED AT DIFFERENT TIMES IN A DIURNAL CYCLE

THE downy mildew of grapes caused by *Plasmopara viticola* (B and C) Berl. and DeT. is a serious disease in peninsular part of India. For effective chemical control of this disease it is necessary to know the germinability of sporangia collected at different times of the day and also when sporangia with the highest germination percentage are produced. In order to obtain this information, grape leaves showing downy mildew growth were gently tapped at 4 p.m. to dislodge the preformed sporangia. From such leaves the sporangia were collected in sterile water by using a soft brush at 2 hr–intervals from 6 p.m. to 4 p.m. the following day. Sporangial suspensions containing 2,00,000 sporangia per ml were prepared. One drop of the suspension was placed in a sterile concavity microslide and incubated in moist chamber in darkness at 30° C. The number of sporangia germinated was counted at hourly intervals for seven hours. From the total number of sporangia in a microscopic field, the per cent of sporangia germinated was calculated. The results are presented in Table I.

TABLE I

Germination of Plasmopara viticola *sporangia collected at different times*

Time of collection	Per cent of Sporangial germination—hr after incubation						
	1	2	3	4	5	6	7
6 p.m.	..	..	..	..	..	..	..
8 p.m.	5·7	10·3	12·0	12·9	13·2	13·4	13·7
10 p.m.	10·6	20·0	23·6	26·9	28·7	31·0	31·6
12 midnight	16·4	20·8	33·2	37·1	38·2	38·4	38·4
2 a.m.	20·4	37·1	41·9	42·9	47·7	48·7	48·7
4 a.m.	24·0	30·5	31·7	32·1	33·4	33·9	33·9
6 a.m.	22·6	24·5	26·6	29·9	30·0	30·2	30·3
8 a.m.	..	5·7	6·3	7·0	8·1	8·1	8·3

C.D. (P = 0·05) between times: 1·8
,, ,, Hours: 1·8
,, ,, Time × Hours interaction 4·7

The data revealed that the sporangia collected from 10 a.m. to 6 p.m. did not germinate even after seven hours. However, sporangia collected from 8 p.m. to 6 a.m. germinated within an hour. Sporangia collected at 8 a.m. germinated only after two hours of incubation. Among the sporangia produced during different times of the day, those collected at 2 a.m. gave the highest percentage of germination. The sporangia collected after that showed a drop in the percentage of germination and reached the lowest percentage when collected at 8 a.m. The differences in mean sporangial germination between different times of collection were significant. The mean sporangial germinations after seven and five hours of incubation were on par.

The data indicated that the sporangia were short lived and did not survive exposure to sunlight at the temperature above 30° C prevailing at Coimbatore. Gregory[3] studied the factors influencing sporangial germination. Cuboni[1] and Faes and Tonduz[2] reported that sporangial germination was prevented by intense light. Viala[4] observed severe incidence of downy mildew in France under conditions of reduced sunlight. High light intensity was attributed as a cause for the absence or rare occurrence of this disease in Southern Greece[5]. The maximum germinability of sporangia collected at 2 a.m. seems to serve a dual purpose in perpetuation of the fungus-avoidance of loss of viability due to sunlight and ensuring infection by making use of water film available on leaf surface for germination during the cool night of diurnal cycle.

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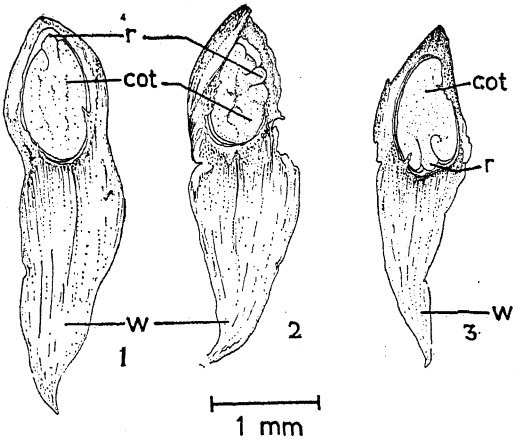
Coimbatore 641 003, *January* 21, 1975.

---

1. Cuboni, J., *Bot. Critt. in Parma*, 1887, p. 91.
2. Faes, H. and Tonduz, P., *Annuaire Agric. de la Swisse*, 1925, p. 657.
3. Gregory, C. T., *Phytopathology*, 1912, 2, 235.
4. Viala, P., *Comptes Rendus Acad. d'Agric. De France*, 1932, 18, 966.
5. Sarejanni, J. A., *Ann. Inst. Phytopath. Benaki*, 1951, 5, 53.

REVERSION OF EMBRYO IN A SEED OF PLUMERIA*

THE seeds of *Plumeria rubra* L. are long (2·6–4–8 cm), flat, slightly hard and dull brown in colour with a large (0·9–3·1 cm) chalazal papery wing. Normally the embryo root is situated towards the micropylar end and the plumule towards the chalazal side (Fig. 1), but on exactly reverse condition (radicle towards chalazal wing side and plumule towards micropylar side) was observed during the dissection of the seeds (Fig. 3). It was quite interesting and created a curiosity to dissect more seeds and by chance an intermediate stage was also found. In this case the embryo root was situated laterally (Fig. 2).



FIGS. 1–3. Seeds of *Plumeria rubra* forma *acutifolia*. Fig. 1. A normal dissected seed with radicle towards micropylar side. Fig. 2. Abnormal dissected seed with laterally placed radicle. Fig. 3. Abnormal dissected seed with radicle towards chalazal side. (*cot*, cotyledons ; *r*, radicle (Embryo root) ; *W*. wing.).

The cases of development of Embryo other than the egg in an embryo sac were reported by many workers (Maheshwari, 1950). But here it is interpreted that there is reversion of the entire embryo through the intermediate stage during the early development of the embryo.

The author is indebted to Prof. V. Puri, Dr. Y. S. Murty (Meerut University) and Sri. S. K. Agarwal (M.S. College, Saharanpur) for their valuable suggestions and to the Ministry of Education, Government of India, for financial help during the tenure of which this work has been done.

Department of Botany, T. S. CHAUHAN.
M.S. College,
Saharanpur, *August* 2, 1975.

---

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1. Maheshwari, P., *An Introduction to the Embryology of Angiosperms*, McGraw-Hill, New York, 1950.

'ROSETTE HABIT'—AN INDUCED PHYSIOLOGICAL MUTATION IN RAGI* OR FINGER-MILLET (*ELEUSINE CORACANA*, GAERTN.)

SPONTANEOUS mutants with a rosette habit have occurred in maize, sweetcolover, peas, *Lolium perene* and others. A study of such mutants resulted in establishing that the deficiency of the growth hormone Gibberellic acid (GA_3) or a similar substance, as responsible for such a habit. Many long-day plants prolong their vegetative phase forming a rosette, under short day conditions ; but the application of GA_3 to such plants induces stem elongation followed by flowering. Induced mutants with a rosette habit and which turn towards normal after GA_3 application have been reported in groundnut[1] and in barley[8]. Induced mutants with a rosette habit have been reported in Cowpea[4] and Cotton[2]. An induced mutant with rosette habit in finger millet (*E. coracana*) was isolated and its response to GA_3 is reported here.

Material and Methods

Treatment with gamma rays and ethyl methane sulphonate and growing of M_1 and M_2 generations are described elsewhere[7]. The mutant plants isolated in the M_2 generation at a month's stage were grown in pots. A spray of gibberellic acid (GA_3) solution at concentrations of 0, 20, 40, 60 and 80 ppm was given to them. Four pots per concentration served as four replications. The stem elongation was measured (from soil surface to the tip of the longest leaf) at the end of the week after GA_3 application. After the first week, spraying of GA_3 solution was continued with 40 ppm concentration only, till the plants had completed their flowering. At this stage plants in 40 ppm treatment were divided into two lots. For one of the lots (A group) spray was stopped for two weeks, while the other lot (B group) received the spray at weekly intervals regularly. Measurements of stem elongation were made before each application of GA_3. The seeds from these GA_3 treated plants were sown in their M_3 generation for confirming the mutant behaviour.

Results and Discussion:

The mutant was recognised as a distinct deviant from the seedling stage with crowded, short, thick and dark green leaves (Fig. 1). Its response to GA_3 application as measured by stem elongation at the end of first week after application is presented in Table I. The data reveal that increase in the height after GA_3 application to be highly significant. Among the treatments the treatment 40 ppm has shown the maximum response. Hence, further treatment with GA_3 was given only at 40 ppm concentration.

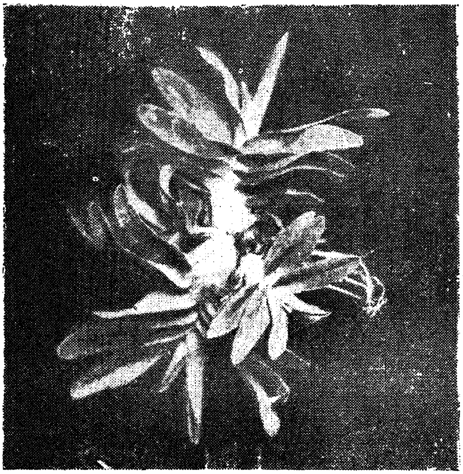

FIG. 1

TABLE I

GA_3 treatments in ppm	0	20	40	60	80
Mean height (in cm)	2·65	6·48	9·48	8·02	4·60
C.D. at 5% level			4·12		
1% level			5·76		

Repeated application was made on the basis of previous reports as in alfa-alfa[3]. The mutants required repeated application of GA_3 for their continuous growth. Treated–A group (Table II) which had a gap of two weeks after the initial application produced little growth after the initial response, whereas, the other treated group with the gap of only one week showed increased growth. These observations confirm the previous reports as mentioned above. The deviants after treatment with GA_3 attained almost normal appearance (Fig. 2) and flowered at a similar period as that of normal (non-mutant parent). The mutant without treatment with GA_3 flowered as late as three months, and the inflorescence was extremely compact and almost hidden in the leaf whorl. Some seed-set was observed in the treated mutants and none in the untreated.

TABLE II

	13th day	18th day	25th day	32nd day	39th day
Control	4·91	4·63	7·33	7·33	7·33
Treated A	16·20	16·20	16·20	157·50	335·00
Treated B	15·50	15·50	276·25	310·00	395·00

Treated A—Gap of two weeks before 2nd application of GA_3

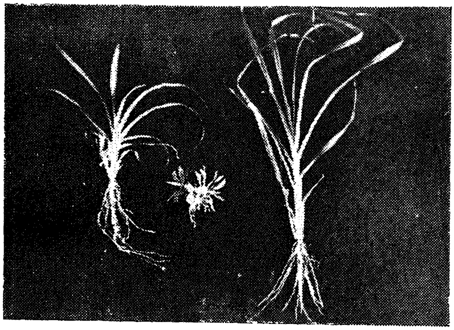

FIG. 2

The seeds from these deviants were carried to the M_3 generation and found to breed true to their type. This indicated the normal synthesis of GA_3 or similar growth substance necessary for the growth of the plant has been affected due to mutation. The M_1 plants resembled normal plants and gave rise to the mutants only in the M_2 generation. In the M_2 generation the segregation ratio varied from 15% in 10 kr treatment to 19% in 40 kr treatment. This is closer to the expected ratio of 20% for a monogenic ratio, after accounting for deficit of recessives due to 'diplontic selection', etc. Hence the mutation involved is recessive to the normal unlike the groundnut mutant[1] which is dominant with variable penetrance and expressivity. However, the barley mutant[8] is similar to the one reported here.

The mutation was observed in three (10, 20 and 40 kr) of the six (0, 10, 20, 30, 40 and 50 kr) doses studied in X-rays, while in no EMS (Ethylmethane sulphonate) treatments, it was observed. Amongst the previous reports the rosette mutant in groundnut has been induced in dES (diethyl sulphate) treatments while that of barley in treatments with X-rays. Ionising radiations are known to destroy the biological activity of auxins[6] (Skoog, 1935) and also that of GA_3[5]. However, the rosette mutants represent destruction of GA_3 activity induced as a genetic change.

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1. Ashri, A., *Mutation. Res.*, 1970, 9, 473.
2. Natarajan, P. T., unpublished M.Sc. (Agri.) dissertation, submitted to Madras University, 1968.
3. Pauli, and Sarenson, *Crop. Sci.*, 1961, 1, 269.
4. Prabhakara Rao, unpublished M.Sc. (Agri.) dissertation, submitted to Madras University, 1968.
5. Sideris, E. G., Eleinhofs, A. and Nilan, R. A., *Radiation Botany*, 1969, 9, 349.
6. Skoog, F., *J. Cell and Comp. Physiol.*, 1935, 17, 455.
7. Sreekantaradhya, R. and Madhava Menon, P., *Madras Agric. J.*, under publication, 1975.
8. Tsuchiya,, T., *Proc. FAO/IAEA Symposium*, Washington, 1969.

ENDOSPERM EMBRYOIDS IN CULTURES OF *NIGELLA DAMASCENA*

THE endosperm has been generally considered morphogenetically inactive, and even dead[1]. But the recent reports of callus formation and of organogenesis in endosperm cultures[2] lend experimental evidence to the morphogenetic potentiality of the endosperm. Nonetheless, our survey shows that the data are limited to 21 species and points to an extensification of morphogenetic studies on endosperm in angiosperms. Our work reports embryoid formation *in vitro* from endosperm tissue of *Nigella damascena* Linn. (Ranunculaceae).

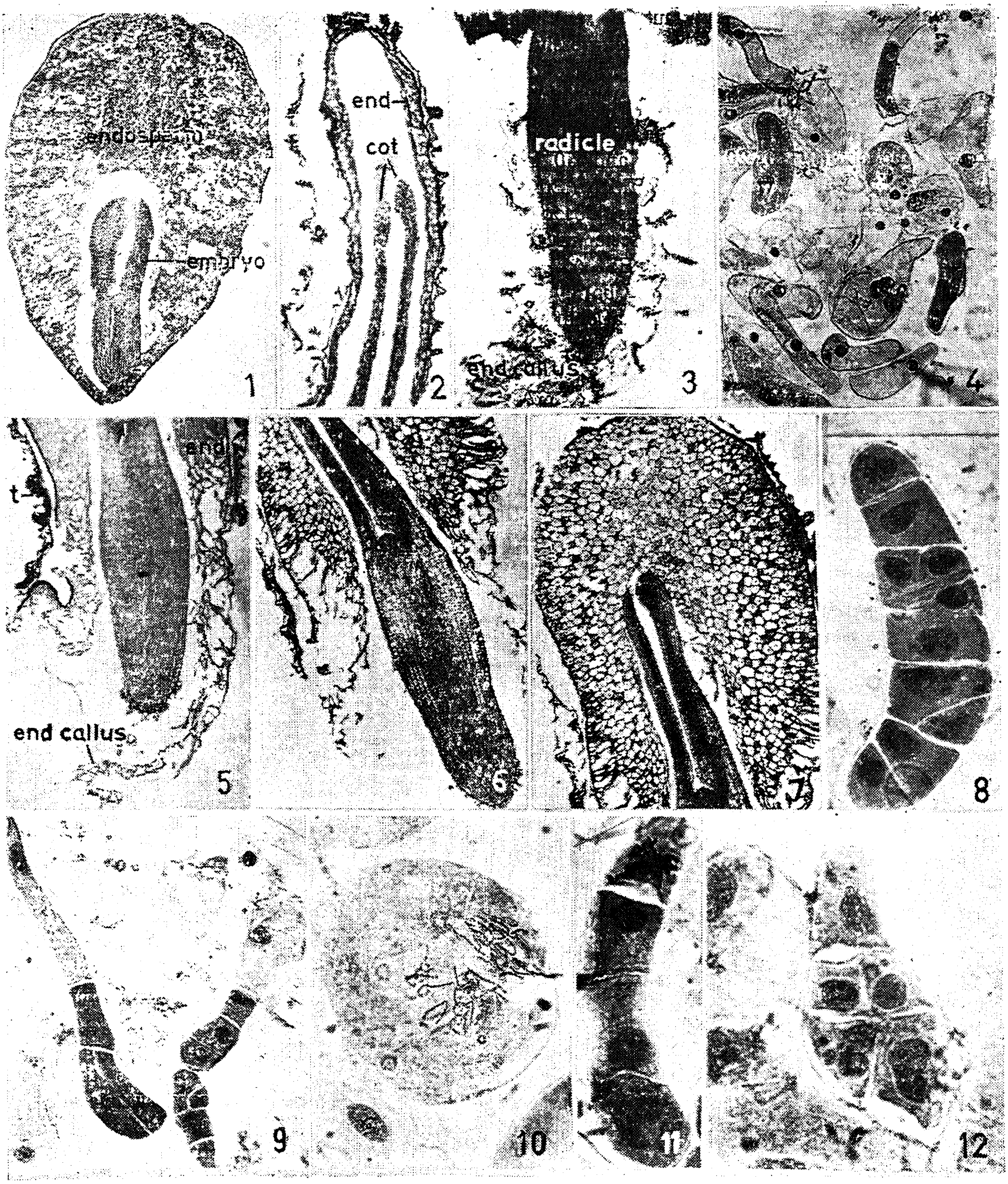
Mature seeds of *Nigella damascena* were obtained from the market. After surface-sterilizing in rectified spirit and nascent chlorine water the seeds were sown individually on Murashige and Skoog's medium containing 3% sucrose and jelled with 0·8% agar (control). The effect of 2,-4-dichlorophenoxyacetic acid (2, 4–D) on seed cultures was studied. In all 192 cultures were raised in 2 replicates of 96 cultures each. The cultures were initially maintained in darkness for 10 days at 25° C ± 2°, and then in 75–80 or 3000–3500 lux at culture level.

At maturity the seed contains a dicotyledonous embryo and 25–30 layers of endosperm (Fig. 1). The endosperm cells are densely cytoplasmic and usually uninucleate. Chemical tests showed the endosperm to be uniformly rich in protein and oil glubules.

On the control medium up to 61% seeds germinated during which process the endosperm was consumed (Fig. 2) as *in vivo*. But on the medium supplemented with 2, 4–D (5×10^{-6}, 10^{-5} M) the radicle emerged in 30% seeds and failed to develop into a root; in 14 days a yellowish, rather translucent callus was seen enclosing the radicle (Fig. 3). The callus was so friable that even a gentle squashing made it dissociate to largely single cells, some 2-celled filaments, and a few clumps of less than 10 cells (Fig. 4). The single cells were either densely cytoplasmic or highly vacuolated. In cell content and staining properties the 2-celled filaments resembled the densely cytoplasmic single cells, indicating that the latter were potential initials of the filamentous structures.

Invariably the callus developed at the micropylar pole of the seed. It originated from up to 5 peripheral layers of the endosperm; the radicle did not contribute to callus formation (Fig. 5). In cultures in which the callus growth was extensive more and more of the endosperm tissue farther from the micropylar region also became proliferative and, the newly formed endosperm callus fanned out from within the seed (Fig. 6); that part of the endosperm tissue which lay *in situ* also increased as much as by 8 layers (Fig. 7).

By 25 days of culture the endosperm callus further overarched the radicle leaving only its tip



FIGS. 1–12. Morphogenesis of endosperm in seed cultures of *Nigella damascena* (*cot* = cotyledon, *end* = endosperm, *end callus* = endosperm callus, *t* = testa). Fig. 1. L.s. seed at culture, × 20. Fig. 2. L.s. seed above the level of shoot apex (note cotyledons), 28 days after culture on Ms (control); hardly any endosperm is seen persistent, × 18. Fig. 3. Wholemount of veil-like friable endosperm callus around the radicle seen 14 days after seed culture on MS + 2, 4–D 10^{-5} M; the shoulders at the distal end of the radicle are of the ruptured testa, × 21. Fig. 4. Part of squash preparation of endosperm callus 63 days after seed culture; note single cells of 2 types—vacuolated and richly cytoplasmic; no cell clump is seen in this particular field of squash preparation, × 75. Fig. 5. L.s. seed through micropyle upward, 15 days after culture on MS + 2, 4–D 5×10^{-6} M. The radicle has emerged and is histogenetically intact. Observe the overarching

friable endosperm callus and its continuity with the parent endosperm *in situ;* note the extent of the ruptured testa and the level of shoot apex (arrow), × 20. Figs. 6, 7. L.s. seed in 2 parts 14 days after culture on MS + 2, 4–D 10^{-5} M. In Fig. 6 the result of meristematic activity of the endosperm at and above the level of shoot apex is evident by the radial files of endosperm tissue flaring out from the seed. Fig. 7. Shows the remaining part of the same seed; compare with Figs. 1 and 2, Fig. 6, × 22, Fig. 7, × 18. Figs. 8, 9. Filamentous embryoids isolated from endosperm callus 68 and 71 days after seed culture; Fig. 8, × 179, Fig. 9, × 93. Figs. 10, 11. Mitosis in an isolated cell and in a filament, Fig. 10, × 105, Fig. 11, × 351. Fig. 12. Globular embryoid in wholemount of endosperm callus, × 297.

visible. The callus continued to be friable even 60 days after culture. It presented fewer single cells but an increased number of filamentous structures (Figs. 8, 9) which resembled the early stages of embryogeny of this species *in vivo*[3]. Mitoses were observed in both—densely cytoplasmic single cells and filamentous embryoids (Figs. 10, 11). Although the chromosome number could not easily be determined, counts made from the dividing single cells and filaments showed it to be nearly the triploid number characteristic of the endosperm tissue ($n = 6$ for *Nigella damascena*[4]). Embryoid formation occurred in 100% seed cultures in which the endosperm had callused. In ageing cultures the embryoids differentiated copiously and some reached the globular stage (Fig. 12). Although a given population of single cells could not be kept under continuous observation, the close cytological similarity between embryoids and densely cytoplasmic single cells, and the incidence of mitoses in the latter strongly indicate that the embryoids are of single cell origin.

Reports of successful endosperm culture are sporadic; of 21 species so far studied organogenesis has been achieved in 9 taxa (6 parasitic and 3 autotrophic), and embryoid differentiation in only 3 taxa—*Nigella* (present work), *Ricinus*[5] and *Croton*[6]. Interestingly, in all these 3 taxa the embryoids differentiated up to globular stage only. In view of the rare differentiation of endosperm embryoids, that such embryoids have been obtained in 100% callused cultures and in so copious a quantity in *Nigella damascena* are significant. Further investigations must be directed to examine what makes the endosperm more recalcitrant than other tissues to organogenesis in general and to embryoid differentiation in particular.

We thank the Council of Scientific and Industrial Research, New Delhi, for a research fellowship to one us (MS). We express our sincere appreciation to Professor H. Y. Mohan Ram, for facilities, and to Dr. M. R. Vijayaraghavan, for photography.

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---

1. Esau, K., *Plant Anatomy* (2nd edn.), John Wiley and Sons, New York, London, Sydney, 1965, p. 617.
2. Johri, B. M. and Srivastava, P. S., *Z. Pflanzenphysiol.*, 1973, 70, 285.
3. Vijayaraghavan, M. R. and Marwah, Kanwal Nain, *Phytomorphology*, 1969, 19, 147.
4. Darlington, C. D. and Wylie, A. P., *Chromosome Atlas of Flowering Plants,* George Allen and Unwin Ltd., London, 1961, p. 19.
5. Satsangi, Asha, and Mohan Ram, H. Y., *Phytomorphology,* 1965, 15, 26.
6. Bhojwani, S. S., *Ibid.,* 1966, 16, 349.

BACTERIAL BLIGHT OF 'FOUR-O'CLOCK'—A NEW DISEASE IN INDIA

A SEVERE blight disease was observed to be prevalent on bushes of 'four-O'clock' (*Mirabilis jalapa* L.) during monsoon months of 1970 and 1971, in Simla (1800–2100 m.a.s.l.) and Almora (1500–1650 m.a.s.l.), respectively. In nature, the disease was manifested as foliar spotting and blighting. The spots were brown to dark brown, circular or irregular and measured about 0·1–0·5 cm in diameter. The blight symptoms appeared as marginal necrosis on leaves, extended inward rapidly along the veins under favourable weather conditions (Fig. 1). Severely blighted leaves were shed-off.

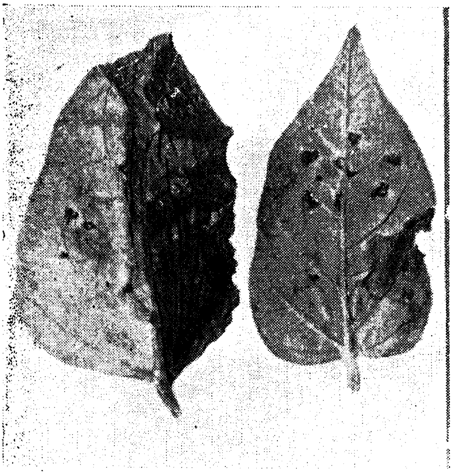

FIG. 1. Symptoms of bacterial blight on *Mirabilis jalapa* leaves.

In advance cases the stem was also invaded through petiole of blighted leaves, turned brown and shrank in girth. Ultimately, plants were destroyed completely.

Microscopic examination of affected plant parts suggested bacterial etiology of the disease. In repeated isolations, a yellow pigmented bacterium, growing as circular, raised, smooth, convex, glistening and butyrous colonies with entire margins on NA, was obtained. Typical symptoms of the disease were reproduced on young healthy host plants within 15 days by spray-inoculating an aqueous suspension of the bacterium.

The phytopathogenic organism was Gram-negative, rod-shaped, mostly single, capsulated, non-sporing, non-acid fast and motile with single polar flagellum. The biochemical and physiological characteristics of the bacterium were: strictly aerobic; utilized glucose oxidatively; gelatin liquefied; starch and aesculin hydrolysed; lypolytic with Tween-80; produced ammonia, hydrogen sulphide and catalase but not urease, tyrosinase and oxidase; nitrate reduction, indole production, Voges-Proskauer and methyl red tests were negative; asparagine was not utilized as a sole source of carbon and nitrogen; growth inhibited by 5% salt in broth, milk peptonized without coagulation; litmus not reduced; produced acid without gas from arabinose, ribose, xylose, fructose, galactose, glucose, mannose, maltose, sucrose, malibiose, trehlose, dextrin, glycogen, starch, glycerol, mannitol but not from rhamnose, lactose, sorbose, raffinose, inulin, erythritol; sorbitol, dulcitol, salicin, α-methyl-D-glucoside and meso-inositol; utilized sodium salts of acetic, citric, lactic, propionic and succinic acids but not of benzoic acid.

The cultural, morphological and physiological properties of the phytopathogenic organism suggested that the bacterium belongs to the genus *Xanthomonas.*

Host range studies revealed that the xanthomonad was capable of infecting *Cyamopsis tetragonoloba, Phaseolus lunatus, P. vulgaris* and *Vicia faba.* The susceptibility of *V. faba* was exceptionally more than other hosts. It was also observed that *X. phaseoli,* incitant of common blight of beans, could not infect *M. jalapa.*

Though the pathogen could infect a few leguminous hosts, it obviously differed from *X. cyamopsidis* Patel, Dhande and Kulkarni[2,3], *X. phaseoli* (Smith) Dowson[4] and *X. vignicola* Burk[1]., in host range and some physiological properties. The incitant of the bacterial blight of 'four-O'clock' is, therefore, identified as a new species and the binomial *Xanthomonas mirabilis* sp. nov. is proposed.

The disease is a new record to science. The extent to which *X. mirabilis* would be threatening the leguminous crops in temperate regions in India needs to be investigated.

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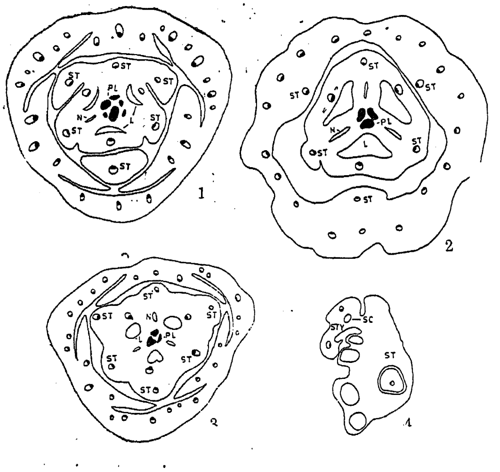
---

1. Breed, R. S., *et al., Bergey's Manual of Determinative Bacteriology,* 7th ed., Baillier, Tindall and Cox, London, 1957, p. 170.
2. Patel, A. J. and Patel, M. K., *Curr. Sci.,* 1958, 27, 258.
3. Patel, M. K., *et al., Ibid.,* 1952, 21, 183.
4. Patel, P. N. and Jindal, J. K., *Indian Phytopath.,* 1972, **25**, 517.

AN INCIPIENT GYNOSTEMIUM IN THE ALOINEAE (LILIACEAE)

During a study on the floral anatomy and morphology of the Liliaceae, an interesting case of fusion of the bases of some stamens with the ovary was observed in *Aloe barbadensis* Mill. To confirm this, a number of species of *Aloe* and also the related genera, *Haworthia* and *Gasteria,* were studied.

The number of stamens in all the species is six and these are arranged in two alternating whorls of three each. In *A. barbadensis,* four to five stamens are united with the base of the ovary for a short length (Fig. 1). In *Haworthia limifolia* Marloth.,



Figs. 1–4. Fig. 1. *Aloe* **barbadensis.** Fig. 2. *Haworthia limifolia.* Fig. 3. *Gasteria trigona* ? Fig. 4. *Aloe barbadensis,* a stamen united with the *style* (*L,* loculus; *N,* septal nectary; *PL,* placental bundles. *S.C.,* stylar canal; *ST,* stamen or a staminal strand; *Sty,* style).

the inner stamens are united with the ovary for a greater length, while the outer stamens are adnate to the perianth tube (Fig. 2). In a species of *Gasteria* (*G. trigona* Haw. ?), all the six stamens are adnate to the ovary for a still greater length (Fig. 3). In some of the sixteen studied species of *Aloe, Haworthia* and *Gasteria,* no such adnation was observed, while in a few others one or the other of the above variations were met with. In abnormal flowers of *A. barbadensis* and *H. limifolia,* one of the six stamens (generally an antero-lateral one of the inner androecial whorl) is united with the style as well (Fig. 4).

In some other liliaceous genera, *e.g., Ornithoglossum viride* var. *undulatum,* Buxbaum[1] records coalescence of four of the six stamens with the ovary. In abnormal flowers of *Tofieldia calyculata,* Leinfellner[3] records a similar adnation.

A marked adnation of the stamens with the gynoecium has been recorded for the Apostasiaceae[4]. In the Orchidaceae, the fusion of the staminal and stigmatic whorls has reached an extreme degree, and the gynostemium appears like a central pillar-like structure[6]. These are highly advanced taxa of the petaloid monocotyledons.

The Liliaceae are generally considered as the basic stock for the evolution of many families of the petaloid monocotyledonae[2]. Moreover, this evolution is thought to be along a few, related and parallel, lines[5]. The occurrence of an incipient gynostemium in the Aloineae is, therefore, significant. It indicates that the trend towards the development of a gynostemium does exist in the Liliaceae.

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---

1. Buxbaum, F., *Öst. bot. Z.,* 1954, **101**, 337.
2. Lawrence, G. H. M., *Taxonomy of Vascular Plants,* Macmillan, N.Y., 1951.
3. Leinfellner, W., *Öst. bot. Z.,* 1962, **109**, 395.
4. Rao, V. S., *J. Indian bot. Soc.,* 1969, **48**, 374.
5. Stebbins, G. L. and Khush, G. S., *Amer. J. Bot.,* 1961, **48**, 51.
6. Swamy, B. G. L., *Bot. Mus. Leafl. Harvard Univ.,* 1948, **13**, 61.

BACTERIAL LEAF SPOT OF GARDEN NASTURTIUM IN INDIA

DURING monsoon months of 1969, a severe foliar spotting disease of garden nasturtium (*Tropaeolum majus* L.) was observed in Simla (1800–2100 m. a.s.l.) (Fig. 1). The spots were small, circular, necrotic brown and about 0·5–1·0 mm in diameter. Narrow brown lesions of about 0·5 cm length were also present on petiole and stem. Severely affected plants exhibited premature yellowing and drooping of leaves. Microscopic examination of necrotic tissue of spots suggested bacterial etiology of the disease.

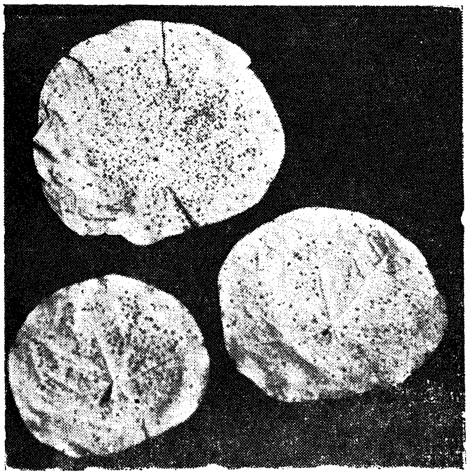

FIG. 1. Bacterial leaf spots on *Tropaeolum majus.*

From fresh spots, a green fluorescent bacterium forming small, circular white, translucent, smooth, flat and glistening colonies with entire margins on NA was isolated. The isolate produced typical syptoms of the disease within 5–7 days when spray-inoculated on young healthy host plants. The pathogenic bacterium which was rod-shaped, Gram-negative, non-spore forming, non-acid fast and motile with 1–4 polar flagella (occasionally bipolar), showed the following physiological characteristics : aerobic; utilized glucose oxidatively; gelatin not liquefied; produced ammonia but not hydrogen sulphide, indole, acetoin and levan, starch not hydrolysed; nitrate not reduced; tolerated 5% salt in broth; no peptonization and coagulation of milk; produced acid without gas from galactose, glucose, sucrose, glycerol, mannitol but not from lactose, maltose and silicin, utilized sodium salts of acetic, citric, propionic and tartaric acids but not of benzoic and oxalic acids.

The bacterium has been identified as a species of *Pseudomonas.* The disease is new for this host in India, *P. aptata* (Brown and Jemieson) Stevens has been reported to produce leaf spots on garden nasturtium and sugarbeet (*Beta vulgaris* L.)[2]. The present isolate, however, did not infect sugarbeet and distinguished itself from *P. aptata* in some physiological and biochemical characteristics also[1]. The differences, therefore, suggest that the *Pseudomonas* sp. isolated from *T. majus* in India is a distinct strain of *P. aptata*.

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New Delhi, *August* 8, 1975.

---

1. Breed, R. S. *et al.*, *Bergey's Manual of Determinative Bacteriology*, 7th ed., Bailliere, Tindall and Cox., London, 1957, p. 122.
2. Elliott, C., *Manual of Bacterial Plant Pathogens*, 2nd ed., Chronica Botanica Co., Waltham, Mass., U.S.A., 1951, p. 56.

BRASSICA CAMPESTRIS VAR. *TORIA*—A NEW HOST FOR *HELLULA UNDALIS* F. (PYRALIDAE: LEPIDOPTERA)

Hellula undalis F. has been reported feeding on cabbage, cauliflower, turnip, radish, at times on mustard, seedings of egg plants, spinach, beet and cabbage. The larvae eat the growing point and burrow in the stem and larger leaves of cabbage[1] and a single plant is seen harbouring as many as 70 larvae[2].

During October 1974, when the crop was 30–45 days old, *B. campestris* var. *toria* (grown at Farm of the Haryana Agricultural University, India) was consumed by *Hellula undalis* F. The larvae feeds the internal content of the stem of the infested plant leaving a tunnel behind and thus reaches up to the root zone. The larvae were collected from the field and reared in the laboratory up to the adult stage. and were identified as *H. undalis* F. This insect has not so far been recorded on this crop earlier by any worker in India or abroad.

Sincere thanks are due to Dr. T. P. Yadava, Economic Botanist (Oilseeds), for providing necessary facilities and to the Director, British Museum, London, for identifying the insect.

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---

1. Johnston, H. B., *Bull. Welcome Trop. Res. Lab. Sudan Govt. Ent. Sect.*, 1930, 31, 67.
2. Rawat, R. R., Rathore, Y. S. and Saxena, D. K., *Indian J. Ent.*, 1968, 30, 235.

THE EFFICACY OF COMBINATION TREATMENT IN PROMOTING MEIOTIC STABILITY OF HEXAPLOID TRITICALE (*TRITICALE HEXAPLOIDE* LART.)

TRITICALE, the first man-made cereal, though has got certain positive points over more conventional wheat, as yet has failed to be commercialized, the main drawback being poor seed-setting due to meiotic instability. Many scientists, who are now working to improve upon the drawbacks of this cereal, have also used mutation breeding as a possible method[1,2]. The present communication emphasizes the use of combination treatment, *i.e.*, treatment with two mutagens in combination, for the achievement of said goal.

For this purpose, seeds of three promising strains of 6 × triticales (Arma 157, PC 201, and BC 223) were subjected to three mutagen treatments—(*a*) 5 Kr, Gamma rays, (*b*) 0·15% Ethylmethane sulfonate, and (*c*) 5 Kr, Gamma rays + 0·15% EMS in combination. For irradiation, dry seeds were used and for EMS, post-treatment (8 hours of-soaking in distilled water followed by 12 hours of mutagen treatment) was utilized. In the case of combination treatment, dry seeds were first irradiated followed by post-treatment with EMS, 250 seeds being used per treatment. M_1 generation was raised along with the controls in the field in November 1972. Seeds from M_1 population were collected and M_2 raised therefrom in November 1973. In M_2 twenty plants per mutagen treatment were studied for meiotic stability—the parameters being: (1) Mean univalents per cell in first and second metaphase, (2) mean laggards per cell in first and second anaphase, (3) mean laggards per cell in first and second telophase and (4) mean micronuclei per tetrad cell.

The data on the frequency, of univalents at first and second metaphase are presented in Table I. The number of irregular cells was found to be drastically reduced in all mutagenically treated strains but more so in the case of combination treatment. In each case the total number of cells scored being 50. The same holds true for the appearance of mean univalents per cell, the only exception being Arma 157, where treatment of Gamma rays scores a point over combination treatment. Table II summarizes the data on the frequency of laggards at first and second anaphase. It is evident that the combination treatment is better as compared to either of the two in this case also. The data on the frequency of laggards at telophase one and two has been presented in Table III and that for frequency of micronuclei in tetrad cells in

TABLE I

Frequency of univalents at M_1 and M_2

Treatments →	Control				5 Kr Gamma rays				0·15% EMS				5 Kr Gamma rays + 0·15% EMS			
Strains	No. of Irregular cells		Mean Univalents per cell		No. of Irregular cells		Mean Univalents per cell		No. of Irregular cells		Mean Univalents per cell		No. of Irregular cells		Mean Univalents per cell	
	M_1	M_2	M_1	M_2	M_1	M_2	M_1	M_2	M_1	M_2	M_1	M_2	M_1	M_2	M_1	M_2
Arma 157	45	44	1·7	2·3	34	42	1·3	1·6	38	38	1·5	2·0	33	33	1·6	1·7
PC 201	42	33	1·7	1·5	39	39	2·0	2·0	38	34	1·8	1·7	34	33	1·7	1·5
BC 223	38	33	1·4	1·3	31	28	1·3	1·3	30	38	1·2	1·3	25	22	1·3	1·1

TABLE II

Frequency of laggards at A_1 and A_2

Treatments →	Control				5 Kr Gamma rays				0·15% EMS				5 Kr Gamma rays + 0·15% EMS			
Strains	No. of Irregular cells		Mean Laggards per cell		No. of Irregular cells		Mean Laggards per cell		No. of Irregular cells		Mean Laggards per cell		No. of Irregular cells		Mean Laggards per cell	
	A_1	A_2	A_1	A_2	A_1	A_2	A_1	A_2	A_1	A_2	A_1	A_2	A_1	A_2	A_1	A_2
Arma 157	46	42	3·8	2·0	36	38	3·2	1·4	38	39	2·5	1·7	35	36	2·1	1·6
PC 201	42	43	2·6	2·2	41	36	3·0	1·8	37	38	3·3	2·2	35	38	3·2	1·5
BC 223	39	35	1·8	2·4	27	25	1·3	2·3	36	38	1·5	2·1	23	27	1·3	1·6

TABLE III

Frequency of laggards at T_1 and T_2

Treatments →	Control				5 Kr Gamma rays				0·15% EMS				5 Kr Gamma rays + 0·15% EMS			
Strains	No. of Irregular cells		Mean Laggards per cell		No. of Irregular cells		Mean Laggards per cell		No. of Irregular cells		Mean Laggards per cell		No. of Irregular cells		Mean Laggards per cell	
	T_1	T_2	T_1	T_2	T_1	T_2	T_1	T_2	T_1	T_2	T_1	T_2	T_1	T_2	T_1	T_2
Arma 157	49	44	2·8	2·5	33	35	1·6	1·8	41	40	1·9	2·7	35	37	1·7	2·1
PC 201	37	42	1·4	2·0	39	36	2·0	1·9	35	40	1·7	2·1	34	35	1·5	1·5
BC 223	37	40	1·6	1·9	20	25	1·3	1·6	36	32	1·1	1·6	16	24	1·1	1·3

Table IV. In both the cases mutagen treatment seems to have rendered meiosis more stable.

To further check the validity of this observation, the significance of the data was calculated using

TABLE IV

Frequency of micronuclei in tetrad cells

Treatments →	Control		5 Kr Gamma rays		0·15% EMS		5 Kr Gamma rays + 0·15% EMS	
Strains	No. of Irregular cells	Mean Micronuclei per cell	No. of Irregular cells	Mean Micronuclei per cell	No. of Irregular cells	Mean Micronuclei per cell	No. of Irregular cells	Mean Micronuclei per cell
Arma 157	47	2·1	39	2·0	35	1·7	29	1·3
PC 201	33	1·7	35	1·9	38	1·3	36	1·6
BC 223	35	1·8	24	1·5	37	1·4	24	1·1

TABLE V

The values of χ^2 for each treatment with respect to control in each variety and for every cytological stage under observation

Strains →		Arma 157		PC 201		BC 223	
Treatments	Stages	χ^2	P	χ^2	P	χ^2	P
5 Kr Gamma rays	M_1	7·293	·01	0·585	·40–·50	2·291	·10–·20
	M_2	0·332	·50–·60	0·786	·10–·20	1·050	·40–·50
	A_1	6·774	·01	0·071	·70–·80	6·416	·01–·02
	A_2	1·00	·30–·40	2·750	·05–·10	4·166	·04–·05
	T_1	22·764	0·001	0·199	·70–·80	11·750	·001
	T_2	4·882	·04–·05	2·097	·10–·20	9·890	·01
	TC	5·315	·04–·05	0·284	·50–·60	5·002	·04–·05
0·15% EMS	M_1	3·472	·05–·10	1·000	·30–·40	2·941	·05–·10
	M_2	2·439	·10–·20	0·045	·80–·90	1·214	·20–·30
	A_1	4·962	·04–·05	1·507	·20–·30	0·480	·40–·50
	A_2	0·585	·40–·50	1·623	·20–·30	0·458	·40–·50
	T_1	7·111	·01	0·199	·60–·70	0·054	·80–·90
	T_2	1·190	·20–·30	0·271	·50–·60	3·774	·05–·10
	TC	9·756	·01	1·214	·20–·30	0·198	·60–·70
5 Kr Gamma rays + 0·15% EMS	M_1	8·391	·01	3·508	·05–·10	7·250	·01
	M_2	6·733	·01	0·000	..	4·888	·04–·05
	A_1	7·787	·01	0·766	·05–·10	10·866	·001
	A_2	2·097	·10–·20	1·623	·20–·30	2·716	·05–·10
	T_1	14·583	·001	0·437	·50–·60	17·702	·001
	T_2	3·183	·05–·10	2·782	·05–·10	11·111	·001
	TC	17·763	·001	0·420	·50–·60	5·002	·04–·05

P = Probability of variation of a given value of χ^2.

2×2 contingency table for χ^2, for each treatment with respect to control in each variety[3]. The values of χ^2 calculated have been put in the form of Table V. The results obtained clearly show that for Arma 157 and BC 223, treatment with Gamma rays and combination treatment give significant results. None of the treatments shows any effect on variety PC 201. Apart from this it can be seen that the results obtained with combination treatment are more significant as compared to either of the other two treatments.

Thus we conclude that combination treatment with 5 Kr Gamma rays and 0·15% EMS has a slight edge over either of the mutagens when applied alone as a tool for stabilizing meiosis in Triticale.

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1. Ruebenbavar, T. and Nalepa, S., *Hodowla Roslin*, 1970, **14**, 315.
2. Srivastava, H. K. and Malik, K. P., *Curr. Sci.*, 1974, **43**, 470.
3. Panse, V. G. and Sukhatme, P. V., *Statistical Methods for Agricultural Workers*, ICAR, New Delhi, 1967.

SHORT SCIENTIFIC NOTES

On the Occurrence of *Nymphula responsalis* Walker as a Pest of *Salvinia* sp. in Kerala State

The caterpillar of a pyralid moth *Nymphula responsalis* Walker is being recorded for the first time as a pest of *Salvinia* sp. (the African payal), a serious water weed of Kerala.

The female moth lays 50–100 eggs singly on the leaves which hatch in 5 or 6 days. The first instar larva mines the tender leaves and moults on the 3rd or 4th day. The second instar caterpillar makes a small flat leaf-case by bringing together bits of leaves. The caterpillars remaining inside the floating cases defoliate the plants and change the cases frequently to suit their size. Defoliation of the plants at the flat phase will impair the floating balance and the attacked plants sink and decay.

The larval period is about 21 days and the full growth larva is about 15 mm long. The final instar spins a silken cocoon inside the leaf case and remain submerged attached to the nodes of the plant on pupation. Pupal period lasts for 5 to 6 days. The adult is a small brown moth with white wavy lines on both the wings. The moths mate within a day of their emergence and start laying eggs on the 3rd night continuing it for 3 days.

The caterpillar is observed to feed on other water weeds like *Pistia stratiotes, Nymphaea* sp., *Azolla* sp. and *Marsilea* sp. besides *Salvinia.*

The author is grateful to the Director, British Museum, London, for identifying the specimen and to Dr. M. R. G. K. Nair, Professor of Entomology, Kerala Agricultural University, for encouragement, critical reading and suggestions.

The author is thankful to Mr. P. J. Tomy, Research Officer (Rice), Moncompu, for facilities provided and his keen interest in the work.

Kerala Agricultural University,
Rice Research Sub-Station,
Moncompu,
August 18, 1975.
P. C. VERGIS.

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1. Bennet, F. D., "Investigations on the insects attacking the aquatic ferns *Salvinia* sp. in Trinidad, and Northern South America," *Proc. Southern Weed Conference*, 1966, **19**, **497.**

Maize Borer [*Chilo partellus* (Swinhoe)] as Carrier of *Erwinia carotovora* var. *zeae*, the Causal Agent of Bacterial Stalk Rot of Maize

Bacterial stalk rot of maize incited by *Erwinia carotovora* var. *zeae* is a serious disease of maize and causes heavy losses under favourable conditions. In nature the symptoms usually appear at flowering stage and are mostly confined to the basal internodes. However. in some cases the infection is also observed at the upper portion of the stalk usually along with the borer attack. Therefore, experiments were conducted to study the role of maize borer [*Chilo partellus* (Swinhoe)] in transmission of the stalk rot pathogen.

Healthy larvae were fed on 48 hour old bacterial culture growing on nutrient agar in Petri plates while in control the larvae were fed on uninoculated plates of nutrient agar. After 1 hour of feeding, the larvae were transferred into the sheaths of 45 days old healthy maize plants grown in pots.

A single larva was transferred per plant and the sheath containing the larva was tied with the thread to avoid its escape.

The typical symptoms of bacterial stalk rot along with borer damage were observed in 20–25% of the inoculated plants. In 25% of the plants the larvae died inside the sheath without causing any damage while in the remaining inoculated and control plants only borer damage was noticed. The symptoms of the disease in these plants appeared after 7–10 days as compared to 4–6 days in the case of plants directly inoculated with the pure culture of the bacterium and this delay in symptom expression may be due to the low level of inoculum carried by the single larva. Successful transmission of the bacterium was confirmed by isolation of the pathogen from the infected plants. Indication was also obtained that the larvae may carry the bacterium internally; however, it requires further confirmation.

The authors express their gratefulness to Dr. G. S. Sandhu, Entomologist (Research), P.A.U., Ludhiana, for supplying the larvae of maize borer.

Department of Plant Pathology, B. S. THIND.
Punjab Agricultural University, NIRMALJIT SINGH.
Ludhiana, *January* 8, 1975.

A New Host Record of the Vegetable Mite, *Tetranychus cucurbitae* Rahman and Sapra

The vegetable mite, *Tetranychus cucurbitae* Rahman and Sapra is known to feed on several host plants. Among these, cucurbitaceous plants, tomato, holyhock, groundnut, jasmine and peach are important (Sidhu and Gurdip Singh, 1972). In the months of June and July, 1975, the vegetable mite, *T. cucurbitae* was found infesting the exotic weed, *Parthenium histerophorus* L. (Compositae) on the farm of Agricultural College, Dharwar, Karnataka. This was confirmed by allowing the mites to feed on the parthenium plants in the green house. All the stages of the mite (eggs, nymphs and adults) were noticed. The mites produced characteristic white specks on the leaves due to the feeding activity.

Dept. of Entomology and Zoology, PUTTASWAMY.
College of Agriculture, M. C. DEVAIAH.
Dharwar 580 005, *July* 31, 1975. M. R. RANGASWAMY.

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1. Sidhu, A. S. and Gurdipsingh, "Laboratory studies on the host range of the vegetable mite, *Tetranychus cucurbitae* Rahman and Sapra," *J. Res.*, 1972, 9, 513.

ANNOUNCEMENTS

Award of Research Degrees

Karnatak University, Dharwar, has awarded the Ph.D. degree in Zoology to Shri S. A. Nevagi, Shri Shivappa Sengappa Rodgi, Shri Sharabanna Doddappa Amoji, Shri Y. F. Neelgund, (Miss) Angara Venkata Narasubhai.

Karnatak University, Dharwar, has awarded the Ph.D. degree in Geology to Shri R. S. Hanagodimath, Shri Gothe Narasimha Naryan.

Karnatak University, Dharwar, has awarded the Ph.D. degree in Anthropology to Shri B. G. Halbar, (Mrs.) Prabha V. Mahale.

The M.S. University of Baroda has awarded the Ph.D. degree in Physics to Shri Maulesh Prataprai Maru; Ph.D. degree in Chemistry to Shri Rasiklal Amulakhbhai Vora, Kumari Purnima Chandrakant Parikh, Shri Kanaiyalal Vanmalidas Masrani; Ph.D. degree in Botany to Shri Satishchandra Jagmohandas Chokshi; Ph.D. degree in Biochemistry to Kumari Shailaja Dattatraya Telang; Ph.D. degree in Geology to Shri Ochhavlal Krishnalal Shah; Ph.D. degree in Zoology to Shri Mahmod Ali Khan; Ph.D. degree in Microbiology to Kumari Freny Homi Doctor; and Shri Jitendrakumar Dhirubhai Desai; Ph.D. degree in Pharmacology to Shri V. S. R. Krishnamurty.

Osmania University, Hyderabad, has awarded the Ph.D. degree in Physics to Shri T. Bhima Sankaram; Ph.D. degree in Chemistry to Shri Satyanarayana Reddy Padala; Ph.D. degree in Astronomy to Shri Arun Potdar; Ph.D. degree in Geophysics to Shri I. B. Rama Prasada Rao, Ph.D. degree in Genetics to Kumari A. Padma; Ph.D. degree in Geology to Shri A. Suryaprakash Rao; Ph.D. degree in Zoology to Shri Syed Badrul Hasan and Smt. Ratnamala.

Tamil Nadu Agricultural University, Coimbatore, has awarded the Ph.D. degree in Agriculture to Shri Abi Cheeran, Shri S. R. Subramanian, Shri M. S. Venugopalan, Shri V. T. Sundaramurthy, Shri U. S. Krishnamurti.

Utkal University, Bhubaneswar, awarded the Ph.D. degree in Mathematics to Shri Sarat Kumar Nayak; Shri Jayanarayan Panda; Ph.D. degree in Physics to Shri Lambodar Prasad Singh; Ph.D. degree in Chemistry to Shri Nrusingh Charan Misra, Shri Nimai Charan Naik, Shri Biren Kumar, Shri Dinabandhu Misra; Ph.D. degree in Botany to Shri K. C. Panda, Shri S. B. Singh, Shri R. Stephen Vinaya Rai; Ph.D. degree in Geology to Shri K. N. Sahu; Ph.D. degree in Medicine to Dr. S. M. Haque.

REVIEWS AND NOTICES OF BOOKS

"Collected Papers of Sir H. Jeffreys" (Vol. 3), Gravity, (Gordon and Breach, London and N.Y.), 1974. Pp. 38 + 661. Price £ 19·60.

For more than sixty years Sir Harold Jeffreys has been publishing his findings on an amazingly large variety of subjects ranging from mathematical physics and probability to astronomy and geophysics. The last topic represents probably his longest and most active association, dating back to as early as 1915. Few scientists in the world have held the field with meaningful impact for so long a time. Indeed, even half the period would be counted as a long enough innings for a good batsman in any branch of science, especially if such a science is to be as abstract as theoretical physics. Sir Harold seems to have surpassed the stamina of most of his contemporaries in the respect. His collected papers in six distinct volumes are a living testimony to his amazing power of giving mathematical shape to interesting physical ideas which he has been offering to the world in generous measure for more than half a century.

The present volume (No. 3) dealing with "gravity" represents a collection of papers on ideas which have earned him considerable fame. This volume classifies his papers chronologically in each of the four broad areas of his activity in this field, *viz.*, (*i*) theory of gravity, (*ii*) analysis of gravity and dynamical astronomy, (*iii*) strength of the earth, and (*iv*) the effect of variation of latitude on bodily tide : Nutation.

Going through the pages of this volume one feels impressed with the versatility of ideas and their effective description which have been manifest in the author's writings from "classical" days to the "modern" times. These papers offer some insight into the reasons for the popularity of some of Sir Harold's books on the subject, (especially the *'Earth'*, *'Earthquakes and Mountains'*, *'Cartesian Tensors'* and *'Asymptotic Expansions'*) which not only summarize the results of his original investigations comprised in this collected volume, but frequently go beyond them. Indeed some of these books, which have already become classics in their own right, make one wonder if this exercise of collecting his original papers all over again at this time was at all worth while. It is difficult to find a satisfactory answer, but perhaps these books cannot quite substitute for the historical perspective that a chronological sequence of these classified papers offers to the interested reader. The archieval value of these papers, which must be the privilege of only a chosen few, is justified in the present case by the magnitude of the impact, Sir Harold's contributions to mathematical physics and geophysics have made on several generations of workers in these exciting fields. Lady Jeffreys, (herself a distiguished physicist in her own right), who has constantly been by her husband's side, must be congratulated on her initiative in compiling these classified papers for the *benefit of posterity.*

A. N. MITRA.

Elliptic Functions. By Serge Lang. (Addison-Wesley, Reading, Massachusetts 01867), 1974. Pp. xii + 326. Price $ 17.50.

In spite of the importance of elliptic curves there was no modern book, till recently, on the subject in English. Professor Lang, has filled the gap.

The book is divided into four parts. The first part gives general results, the modular equation, the modular function field and its automorphisms. The second part treats complex multiplication, Shimura's reciprocity law, *l*-adic and *p*-adic representations of Deuring and applications to Ihara's theory. A knowledge of class field theory is needed here. The third part gives Tate parametrisation and applications to Isogeny theorems and Serre's work on the galois group of division points over number fields. Part four gives Kronecker limit formulae and applications. There are two appendices. The first one reproduces an unpublished paper of Tate where normal forms of elliptic curves and their automorphisms are tabulated. The second appendix treats Tate trace, Cartier operator and Hasse invariant.

The proofs of the important theorems presented are probably the best available. Throughout, the generalisations, the higher dimensions and connections with other branches are brought into the view. In addition, older alternative analytic proofs of some important results are sketched. This book brings together widely scattered material. All these makes the book very valuable. The book makes difficult reading, but an effort is amply rewarding.

V. J. LAL.

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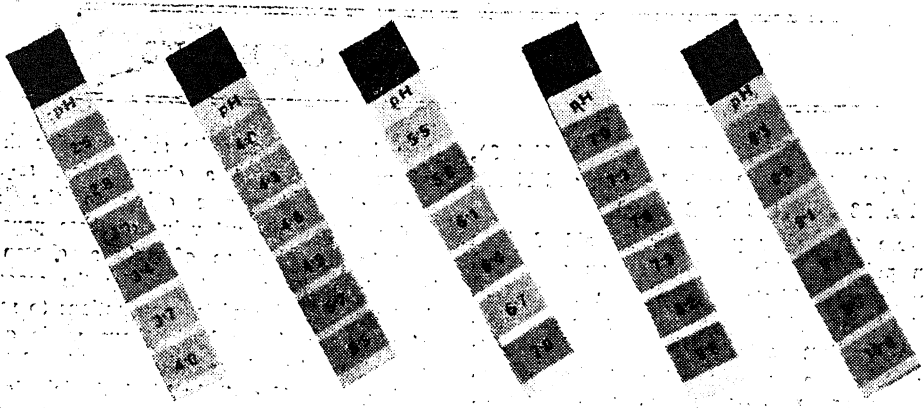

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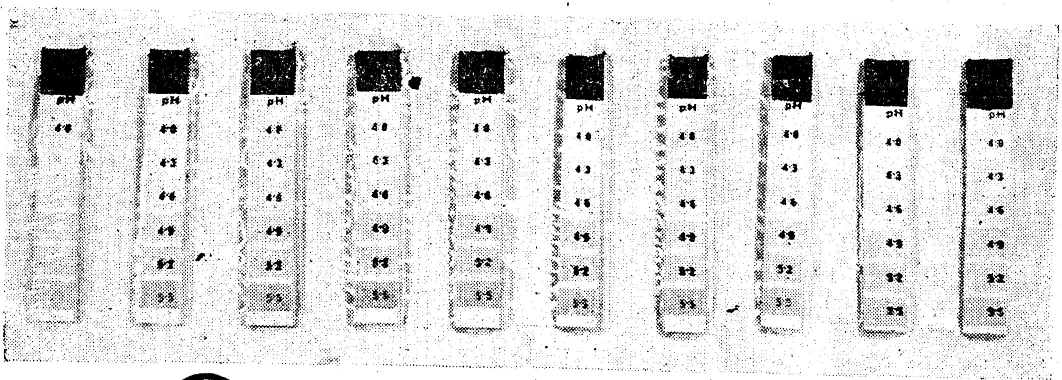

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MATHEMATICAL SOLUTION FOR THE FLOW OF TWO IMMISCIBLE FLUIDS IN A DIPPING CRACKED POROUS MEDIUM WITH MEAN PRESSURE

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ABSTRACT

This paper presents an analytical discussion of the flow of two immiscible liquids in dipping cracked porous medium. The problem has much importance in petroleum technology. An attempt has been made to include capillary pressure effect in the analysis and it is found that the mathematical formulation yields a nonlinear differential equation. This equation of motion has been solved by the perturbation technique to give an analytical expression for the displacing phase saturation distribution under the simplifying assumption that the individual pressure of the two phases may be replaced by their common mean pressure which is regarded as a constant.

INTRODUCTION

THE oil water movement in a cracked porous medium is a problem of petroleum technology and hydrogeology. Many authors have discussed this problem from different viewpoints [Bokserman, Zheltov and Kocheshkov (BZK)[1], Bear and Braester[2], Mattax and Kyte[3] and Verma[4-6]]. Most of the authors except Verma have completely neglected the capillary pressure effect.

In this paper the underlying assumption is made that the individual pressure of the two flowing phases may be replaced by their common mean pressure and expression for the phase saturation is determined. The solution of the nonlinear differential equation has been obtained by perturbation method under the simplifying assumption.

We consider here that water is uniformly injected with a constant velocity into an oil saturation, dipping bed of homogeneous medium traversed by a branched system of cracks varying in orientation. It is assumed that the entire oil on the initial boundary ($x = 0$) is displaced through a small distance due to the impact of injected water.

The main interest of the present investigation is to obtain an analytical expression for phase saturation when the porosity and permeability of the medium may be regarded as constant. The mathematical solution of the nonlinear differential equation of the flow of two immiscible fluids may be obtained under certain special conditions.

2. MATHEMATICAL FORMULATION OF THE PROBLEM

The seepage velocity of oil (V_0) and water (V_w) may be written by Darcy's law as

$$V_0 = -\frac{K_0}{\delta_0} K \left(\frac{\partial p_0}{\partial x} + \rho_0 g \sin \alpha\right), \qquad (2\cdot1)$$

$$V_w = -\frac{K_w}{\delta_w} K \left(\frac{\partial p_w}{\partial x} + \rho_w g \sin \alpha\right). \qquad (2\cdot2)$$

The equation of continuity for the flow of water and oil in the cracked porous medium may be written as

$$P \frac{\partial S_w}{\partial t} + \frac{\partial V_w}{\partial x} + \phi\,[T - \tau(u)] = 0, \qquad (2\cdot3)$$

$$P \frac{\partial S_0}{\partial t} + \frac{\partial V_0}{\partial x} - \phi\,[T - \tau(u)] = 0 \qquad (2\cdot4)$$

where $\phi\,[T - \tau(u)]$ is the impregnation function of the crack system. It is the amount of displacing liquid entering the blocks in an elementary volume of the seam and is analytically defined, following Mattax and Kyte[3] (as in BZK[1]), and Verma[5] by the expression

$$\phi\,[T - \tau(u)] = D\,(T - Rx^2)^{-1/2}, \ \tau \leqslant t$$

$$u = x/L_M, \ T = \theta t, \ R = a/L_M 2$$

$$t = \frac{\delta \cos\theta\, S^2\, (K/m_B)^{1/2}}{\delta_0} \qquad (2\cdot5)$$

$$a = \left(\frac{\pi A}{4q} \cdot \frac{S^2\, \delta \cos\theta\, m_B g_k\, (K/m_B)^{1/2}}{\delta_0}\right)^2$$

From the definition of phase saturation (Scheidegger[7]) we have

$$S_w + S_0 = 1. \qquad (2\cdot6)$$

The capillary pressure (p_c) in a double phase fluid flow is defined to be the pressure discontinuity between the flowing phases across their common interface, and may be written as

$$p_c = p_0 - p_w \qquad (2\cdot7)$$

combining equations (2·1) to (2·4) and using equations (2·6) and (2·7), we get

$$\frac{\partial}{\partial_x}\left[\left(\frac{K_w}{\delta_w} + \frac{K_0}{\delta_0}\right) K \frac{\partial p_0}{\partial x} - \frac{K_w}{\delta_w} K \frac{\partial p_c}{\partial_x} + \left(\frac{K_w}{\delta_w} K\rho_w + \frac{K_0}{\delta_0} K\rho_0\right) g \sin\alpha\right] = 0 \qquad (2\cdot8)$$

Integrating equation (2.8) with respect to x and by simplifying it we get

$$\frac{\partial P_0}{\partial x} = -\frac{V}{(K_w/\delta_w + K_0/\delta_0)K} + \frac{1}{(1 + K_0\delta_w/K_w\delta_0)} \times \frac{\partial P_c}{\partial x} - \frac{\rho_w + \rho_0(K_0\delta_w/K_w\delta_0)}{(1 + K_0\delta_w/K_w\delta_0)} \times g \sin\alpha \tag{2.9}$$

where V is a constant of integration, which can be evaluated later on.

The value of the pressure of oil (p_0) can be written as (Oroveanu[8])

$$p_0 = (p_0 + p_w)/2 + (p_0 - p_w)/2 = \bar{p} + p_c/2 \tag{2.10}$$

where $\bar{p}$ is the mean pressure which is constant.

Substituting the value of $\partial p_0/\partial x$ from equation (2.10) into eq. (2.9), we get

$$V = \frac{1}{2}\left(\frac{K_w}{\delta_w}K - \frac{K_0}{\delta_0}K\right)\frac{\partial p_c}{\partial x} - \left(\frac{K_w}{\delta_w}K\rho_w + \frac{K_0}{\delta_0}K\rho_0\right) g\sin\alpha \tag{2.11}$$

Combining equations (2.3), (2.9) and substituting the value of V and $\phi\,[T - \tau(u)]$ from equations (2.11) and (2.5), we obtain

$$\epsilon P\frac{\partial S_w}{\partial T} + \frac{\partial}{\partial x}\left[\frac{K_w}{\delta_w}K\left\{\left(\frac{1}{2}\right)\frac{dp_c}{dS_w}\cdot\frac{\partial S_w}{\partial x} + \rho_w g\sin\alpha\right\}\right] = -\frac{D}{\sqrt{(T - Rx^2)}}. \tag{2.12}$$

This is the nonlinear differential equation of motion for saturation of two immiscible fluids in dipping cracked porous medium when the capillary pressure is present.

SOLUTION OF THE PROBLEM

It is very difficult to obtain the exact solution of the equation (2.12) due to the nonlinear terms occurring therein. The underlying assumption is that ϵP is small for cracked porous medium of (BZK) because, in this case, $\epsilon \sim 10^{-4}$ to 10^{-6} and $P \sim 10^{-2}$ to 10^{-3}. Therefore, we obtain here an approximate analytical solution of eq. (2.12) by using a perturbation technique[10] (as in Verma[11]), and regarding ϵP as perturbation parameter (as in Verma[5]). Thus, neglecting $(\epsilon P)\,\partial S_w/\partial T$ at the first step, we get

$$\frac{\partial}{\partial x}\left[\frac{K_w}{\delta_w}K\left\{\left(\frac{1}{2}\right)\frac{dp_c}{dS_w}\cdot\frac{\partial S_w}{\partial x} + \rho_w g\sin\alpha\right\}\right] = -\frac{D}{\sqrt{(T - Rx^2)}} \tag{2.13}$$

Performing the integration with respect to x, we get

$$\frac{K_w}{\delta_w}K\left[\left(\frac{1}{2}\right)\frac{dp_c}{dS_w}\cdot\frac{\partial S_w}{\partial x} + \rho_w g\sin\alpha\right] = -\frac{D}{\sqrt{R}}\sin^{-1}(x\sqrt{R/T}) + F(T) \tag{2.14}$$

where F(T) is the constant of integration which is determined by using the condition

$$\partial S_w/\partial x = 0, \quad \text{at} \quad x = L \text{ for all time} \tag{2.15}$$

Substituting equation (2.15) into (2.14), we get

$$\frac{K_w}{\delta_w}K\left[\frac{1}{2}\frac{dp_c}{dS_w}\cdot\frac{\partial S_w}{\partial x} + \rho_w g\sin\alpha\right] = -\frac{D}{\sqrt{R}}\sin^{-1}(x\sqrt{R/t}) + \frac{K_w}{\delta_w}K\rho_w \times g\sin\alpha + \frac{D}{\sqrt{R}}\sin^{-1}(L\sqrt{R/T}). \tag{2.16}$$

At this stage, we assume for definiteness some specific relationships which are quoted in standard literature.

$$K_w = S_w^3, \quad K_0 = 1 - \alpha S_w, \quad \alpha = 1.11$$

$$p_c = \beta(S_w^{-1} - C)\ (\beta \text{ and C are constants}). \tag{2.17}$$

The first one is due to Scheidegger[7] and the second is given by Mishra[9].

Substituting these values in equation (2.16), we get

$$S_w\frac{\partial S_w}{\partial x} = \frac{2D\delta_w}{\sqrt{R}\,K\beta}[\sin^{-1}(x\sqrt{R/T}) - \sin^{-1}(L\sqrt{R/T}). \tag{2.18}$$

Integrating again the equation (2.18) with respect to x, we get

$$\frac{\beta K\sqrt{R}}{4\delta_w D}S_w^2 = x\sin^{-1}(x\sqrt{R/T}) - x\sin^{-1}(L\sqrt{R/T}) + \sqrt{(T/R - x^2)} + E(T), \tag{2.19}$$

where E(T) is the constant of integration which may be evaluated from the following condition:

$$S_w = 1/\alpha \quad \text{at} \quad x = 0. \tag{2.20}$$

Then combining equations (2.19) and (2.20), we get

$$S_w^2 = 1/\alpha^2 + \frac{\sqrt{R}\,\beta K}{4\delta_w D}[x\sin^{-1}(x\sqrt{R/T}) - x\sin^{-1}(L\sqrt{R/T}) + \sqrt{(T/R - x^2)} - \sqrt{T/R}]. \tag{2.21}$$

The next step in the perturbation method is to find the value of $\partial S_w/\partial T$ from equation (2.21) and substitute it in the first term of equation (2.12),

$$\frac{\partial}{\partial x}\left[\frac{K_w}{\delta_w}K\left(\frac{1}{2}\frac{dp_c}{dS_w}\cdot\frac{\partial S_w}{\partial x} + \rho_w g\sin\alpha\right)\right] = -\frac{D}{\sqrt{(T - Rx^2)}} + \left(\frac{1}{2}\right)\epsilon P\sqrt{\left(\frac{\beta K R^{3/2}}{4\delta_w D}\right)} \times \frac{x^2}{T^{3/2}}. \tag{2.22}$$

Integrating equation (2.22) with respect to x under the condition (2.15), we get

$$\frac{1}{2}\frac{K_w}{\delta_w}K\frac{dp_c}{dS_w}\cdot\frac{\partial S_w}{\partial x} = \frac{D}{\sqrt{R}}[\sin^{-1}(L\sqrt{R/T}) - \sin^{-1}(x\sqrt{R/T})] + \left(\frac{1}{6}\right)\epsilon P \times \sqrt{\left(\frac{\beta K R^{3/2}}{4\delta_w D T^3}\right)}(x^3 - L^3) \tag{2.23}$$

substituting the values of K_w, K_0 and p_c from eq. (2·17) in eq. (2·23) and integrating with respect to x under the condition (2·20), we get

$$S_w{}^2 = (1/2\alpha^2) + \frac{4\delta_w}{K\beta}\Big[\frac{D}{\sqrt{R}}(x\sin^{-1})(L\sqrt{R/T}) - x\sin^{-1}(x\sqrt{R/T}) + \sqrt{(T/R - x^2)} - \sqrt{T/R} + (1/6)\,\epsilon P\sqrt{\left(\frac{\beta K R^{3/2}}{4\delta_w D T^3}\right)} \times (x^4/4 - L^3 x)\Big]. \tag{2.24}$$

This is the required solution for the flow of two immiscible fluids in dipping porous medium.

CONCLUSION

In this paper we have obtained the approximate analytical solution of the flow of two immiscible liquids in a dipping cracked porous mediun by perturbation method, under certain conditions. The underlying assumption is that the individual pressure of two flowing phases may be replaced by their common mean pressure, some specific results (which are quoted in standard literature) for relative permeability, capillary pressure, impregnation function have been considered in the present discussion. The problem has gained much current importance in petroleum technology and hydrogeology. The saturation of the water is determined. We have not included any numerical or graphical illustration due to our particular interest in an analytical solution.

ACKNOWLEDGEMENT

The author expresses acknowledgement to Professor A. P. Verma, Head of Department of Applied Mathematics, S.V. Regional College of Engg. and Tech., Surat, for valuable suggestion during preparation of this paper.

The valuable criticisms and suggestions of the reviewers are gratefully acknowledged by the author

NOMENCLATURE

A = Constant;

C = Capillary pressure constant;

K = Permeability of the medium;

K_w = Relative permeability of water ;

K_0 = Relative permeability of oil;

L = Length of the cylindrical porous matrix;

L_M = Mean block size;

m_B = Porosity of the block;

P = Porosity of the crack system;

p_w = Pressure of the water;

p_0 = Pressure of the oil;

q = Average rate of flow in across the face;

S = Mean specific surface area of the blocks;

S_w = Saturation of the water;

S_0 = Saturation of the oil;

t = Time;

T = Weighted time;

u = Mean coordinate;

V_w = Seepage velocity of water;

V_0 = Seepage velocity of oil;

x = Linear coordinate;

β = Capillary pressure coefficient;

ϕ = Impregnation function

δ = Surface tension;

δ_w = Viscosity of the water;

δ_0 = Viscosity of the oil;

g_k = Saturation of blocks with water at the moment t_k;

ϵ = A complex constant of cracked characteristics and the viscosity of oil;

θ = Angle of wetting.

$\tau \leqslant t$

Those symbols which are defined in the text are not included in this table.

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1. Bokserman, A. A., Zheltov, P. Yu. and Kocheshkov, A. A., *Soviet Phys. Doklady*, 1964, **9**, 285.
2. Bear, J. and Braester, C., *Int. Symp. Fund. Trans. Phenom.*, Porous Media III Haifa, 1969, pp 21.
3. Mattax, C. C. and Kyte, I. R., *Petroleum Engg. J.*, 1962, p. 2.
4. Verma, A. P., *Rev. Roum. Sci. Techn. Mec. Appli.*, 1968, p. 277.
5. —, *Can. J. Phys.*, 1969, **47**, 2519.
6. —, *Proc. Nat. Inst. of Science of India*, 1969, **3**, 35.
7. Scheidegger, A. E., *The Physics of Flow through Porous Media*, University of Toronto Press, Toronto, 1960, **216**, 224.
8. Oroveanu, T., (Schuragerea Fluidelor Multifazice Prin Medii Poroase), Ed. *Acad. Bucharest*, 1966, **324**, 386.
9. Mishra, S. K. and Verma, A. P., *Indian J. of Pure and Applied Mathematics*, accepted for publication.
10. Bellmar, R., *Perturbation Techniques in Mathematics, Physics and Engineering*, Holt, Rinehart and Winston, Inc., New York, 1966, p. 2519.
11. Verma, A. P., *Can. J. Phys.*, 1969, **47**, 319.

FOUR AND SIX CO-ORDINATED HYDRAZONE COMPLEXES OF Mn(II)

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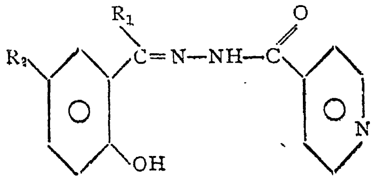
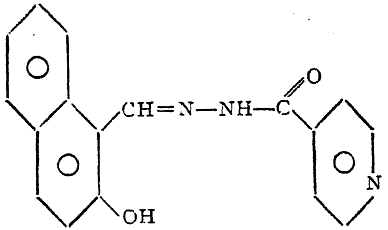
INTRODUCTION

It is well known that the hydrazide residue in the hydrazone can react with metal ions either in the keto or in the enol form[1]. Hydrazones derived from isonicotinoyl hydrazide have been used as chelating agents[2]. 2-OH-5-methyl benzylidene isonicotinoyl hydrazide has been used as an indicator for estimation of Fe(II) by E.D.T.A.[3]. The data on Mn(II) chelates with hydrazone is meagre. It therefore seemed appropriate to study complex formation of manganese (II) with the following hydrazones.





IV

	R_1	R_2		R_1	R_2
I	H	H	V	CH_3	H
II	H	CH_3	VI	CH_3	CH_3
III	H	Cl	VII	CH_3	Cl

EXPERIMENTAL

Mn(II) acetate, isonicotinic acid hydrazide were of reagent grade. Substituted salicylaldehydes and acetophenones were prepared by standard methods[4,5]. Hydrazones were prepared by mixing equimolecular mixtures of hydrazide and aldehyde or ketone in the absolute ethanol. The solid products obtained were recrystallised from ethanol.

The aqueous solution of the metal (II) was mixed with the alcoholic solution of the ligand. The mixture was heated on a water bath for an hour. The precipitated complex was filtered and washed with dilute warm aqueous alcohol to remove the excess ligand and dried over phosphorus pentoxide.

The metal in the complexes was estimated volumetrically with E.D.T.A.[6], after destroying the organic matter by HCl/HNO_3 mixture. Nitrogen was estimated by microanalysis (Table I).

Magnetic susceptibilities were determined on a Gouy balance. Infrared spectra were recorded in the region 4000–650 cm^{-1} on a Perkin Elmer-337 infrared spectrophotometer, using nujol mull. Conductivity was determined on a Philips PR 9500 conductivity bridge cell constant 0·829 cm^{-1}.

RESULTS AND DISCUSSION

Hydrazones I-IV form 1 : 2 complexes whereas V-VII form 1 : 1 complexes with Mn(II).

The complexes are yellow to brown in colour and amorphous in nature. They do not melt upto 300° C, except complex XIV which melts around 250° C with decomposition. They are sparingly

TABLE I

Elemental analysis of Mn (II) complexes of hydrazones

Ligand No.	Complex No.	Empirical formula	Metal analysis Calc. %	Metal analysis Found %	Nitrogen analysis Calc. %	Nitrogen analysis Found %	μ_{eff} at room temp. in B.M.
I	VIII	$Mn(C_{13}H_{10}O_2N_3)_2$	10·28	10·20	15·69	14·94	5·903
II	IX	$Mn(C_{14}H_{12}O_2N_3)_2$	9·67	9·49	14·92	14·29	5·803
III	X	$Mn(C_{13}H_9O_2N_3Cl)_2$	9·11	8·96	13·93	13·36	5·910
IV	XI	$Mn(C_{17}H_{12}O_2N_3)_2$	8·65	9·10	13·28	12·92	5·920
V	XII	$Mn(C_{14}H_{11}ON_3)$	17·80	17·78	13·55	12·96	6·021
VI	XIII	$Mn(C_{15}H_{13}O_2N_3)$	17·00	16·52	12·96	12·91	6·191
VII	IV	$Mn(C_{14}H_{10}O_2N_3Cl)$	15·99	15·69	12·21	12·43	5·981

soluble both in non-polar and polar solvents. Their solutions in DMF do not show measurable conductance, showing their non-electrolytic nature.

The complexes exhibit magnetic moments around 5·90 B.M. indicating the presence of five unpaired electrons in Mn(II). This value is in agreement with the value observed for most of the Mn(II) high spin complexes[7].

In the infrared spectra of ligands I-IV bands due to γ-N—H, γ C=O ; ν C=N and γ C—O (phenolic) are assigned respectively in the regions 3200–3170 cm^{-1}, 1675–1650 cm^{-1}, 1625–1610 cm^{-1} and 1290–1280 cm^{-1}, based on previous assignments[1,8,9,10]. In the complexes VIII-XI γ N—H is retained. Only one strong and broad band is observed in the region 1610–1600 cm^{-1}. It may reasonably be assigned to a combination of γ C=O and γ C=N. The band assigned to phenolic C–O shifts to higher frequency and appears in the region 1310–1295 cm^{-1}.

In the IR spectra of ligands V-VII band due to γ (C=O), γ (N—H), γ (O—H), γ (C=N) and ν (C—O) (phenolic) are respectively assigned in the region 3480–3460 cm^{-1}, 3130–3100 cm^{-1}, 1655–1645 cm^{-1}, 1625–1620 cm^{-1} and 1300–1295 cm^{-1}. In the complexes XII-XIV γ (O–H) and γ (N–H) are absent. The band observed around 1600 cm^{-1} is less intense. This may be attributed to ligands acting in the enolic form. There are two bands in the region 1305–1295 cm^{-1} and these are assigned to phenolic C–O.

The following conclusions can therefore be made. Ligands I-IV react in the keto form and V-VII in the enol form. Nitrogen of C=N and oxygen of C=O coordinate to manganese ; this oxygen in the complexes XII-XIV coordinates in the enolic form. The H of ortho –OH group is replaced by the metal and O is bound to the metal. Thus the complexes VIII-XI have coordination number six. The complexes XII-XIV have 1 : 1 stoichiometry. Their magnetic moments are in agreement with the tetrahedral configuration[11]. Hence the dimeric structure similar to that proposed by Narang *et al.*[1] is suggested for these complexes.

1. Narang, K. K. and Miss Aggarwal, *Inorg. Chim. Acta,* 1974, **9**, 137 (and references there in).
2. Amal, H. and Ergenc, *C.A.*, 1963, **59**, 3515 *d* ; 1964, **60**, 80025 *a* ; A. Doadrio, and Garcia Cargo, *C.A.*, 1966, **65**, 16474 *a*.
3. Gupta, U. S. and Pande, K. K., *Indian J. Appl. Chem.*, 1971, **34**, 106.
4. Duff, J. C., *J. Chem. Soc.*, 1941, p. 547.
5. *Organic Synthesis Collectives,* 1963, **4**, 106.
6. Vogel, A. I., *A Text Book of Quantitative Inorganic Analysis,* Longmann Green (ELBS), 1969.
7. Lewis, J. and Wilkins, R. G., *Modern Coordination Chemistry,* Interscience Publishers, Inc., New York, 1960.
8. Rao, C. N. R., *Chemical Applications of Infrared Spectroscopy,* Academic Press, New York, London, 1962.
9. Clougherty, L. E., Sousa, J. A. and Wyman, G. M., *J. Org. Chem.*, 1957, **22**, 462.
10. Kovacis, J. E., *Spectrochim. Acta,* 1967, **23 A**, 183.
11 Figgis, B. N. and Lewis, J., *Prog. Inorg. Chem.*, 1964, **6**, 37.

VARIATION IN GLYCOGEN, TOTAL CARBOHYDRATES, FREE SUGARS, PROTEINS, TOTAL FREE AMINO ACIDS AND SOME ENZYMES (ACTIVE PHOSPHORYLASE, ACID, AND ALKALINE PHOSPHATASES) DURING EMBRYONIC DEVELOPMENT OF *ANTHERAEA MYLITTA*

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ABSTRACT

Antheraea mylitta during embryonic development shows high acid and alkaline phosphatase activity prior to hatching. The predominance of acid phosphatase activity over that of alkaline phosphatase is noteworthy.

Glycogen initially increases but subsequently decreases by about 16·5% of the initial concentration. Glycogen variation pattern is in tune with that of active phosphorylase activity.

Total carbohydrates and proteins decreased during early stages of development whereafter on 6th day they steadily reattained the initial concentration. Total free amino acids increased with development and exhibited a peak on the third day of development but gradually declined prior to hatching of the first instar larva.

INTRODUCTION

INSECT egg being cleidoic in nature manifests an interesting metabolic pattern. Despite several attempts[1,2], to explore the relationship between some enzymic activities like phosphatases and growth, no definite conclusions could be drawn due to diversity of results obtained in various insects. However, some investigators[3-5], support the view regarding the existence of a definite relationship between the acid and alkaline phosphatase activity and growth and moulting of insects during their life cycle.

Recently, Pant and Nautiyal[6], observed marked utilization of glycogen during embryogenesis of *Philosamia ricini*. In addition, significant correlationship of glycogen degradation to active phosphorylase activity was also observed in this insect during this period. Importance of glycogen as the stored carbohydrate and as a primary fuel reserve in insect eggs is now well known[7-10].

Studies made on embryonic development of *Philosamia ricini*[11] *Schistocerca gregaria*[12], *Sphaerodema molestum*[13] and the grasshopper *Chrotophaga viridifasciata*[14] suggested that yolk reserve proteins are hydrolysed to amino acids and subsequently initiate and get transformed into organ specific proteins required for morphogenesis.

In view of the above it was thought of interest to study the variation in the activity of some important enzymes (acid and alkaline phosphatases and active phosphorylase) and of some metabolites (glycogen, total carbohydrates, reducing sugars, free amino acids and protein content) during embryogenesis of *Antheraea mylitta*, an insect of considerable economic importance in Indian silk industry.

MATERIALS AND METHODS

Antheraea mylitta (Tasar silkworm) cocoons were procured from the Central Tasar Silk Institute, Ranchi, Bihar. After emergence when the female adults started laying, the fertilized eggs were collected, pooled and labelled with the date of oviposition.

300 eggs (weighed, chilled, cleaned with water, ethanol and air-dried) were homogenized with ice-cold glass-distilled water (10 ml) and strained through folds of nylon cloth to remove egg shells. Weight of dry tissue/ml was obtained by heating 1·0 ml of egg homogenate in a tared crucible to constant weight at 80°.

Alkaline and acid phosphate activity were determined with appropriate blanks of buffered sodium β-glycerophosphate substrate of Bodansky[15]. The liberated phosphate was estimated by Sumner's method[16].

Active phosphorylase activity was assayed by the method of Green and Stumpf[17] as modified by Krishnan and Srivastava[18]. Details of the assay mixture were essentially the same as described by Pant and Nautiyal[6]. Glycogen was isolated as described by Weins and Gilbert[19] and estimated by the method of Carrol *et al.*[20], employing anthrone reagent. Total carbohydrates were estimated by the method of Trevelyan and Harrison[21]. Reducing carbohydrates were estimated by the method of Nelson and Somogyi[22]. Total free amino acids and proteins were estimated respectively by employing the method of Yemm and Cocking[23] and Lowry *et al.*[24].

RESULTS AND DISCUSSION

Figure 1 represents the variation in acid and alkaline phosphate activity during embryogenesis of *Antheraea mylitta*. Acid phosphatase activity increases twofold on day 3 and with a subsequent decline on days 4 and 5 depicts the maximum activity on day 6 prior to hatching. Alkaline phosphatase on the other hand, with more or less the same type of variation as its counterpart, reattains the initial activity prior to larva emergance. However, the activity is maintained all along at a lower level than that of acid phosphatase.

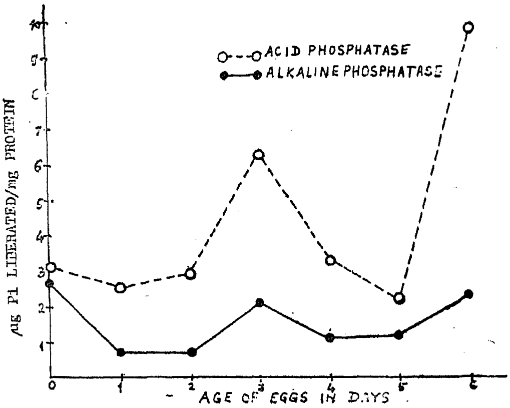

FIG. 1. Variation in acid and alkaline phosphatase activity during embryogenesis of *A. mylitta*.

The predominance of acid phosphatase activity over that of alkaline phosphatase during embryonic development is concordant with that observed in *Bombyx mori*[3] and *P. ricini*[4]. In *B. mori* however, acid phosphatase activity increased throughout embryogeoesis while alkaline phosphatase was inactive during early development[3]. In *Philosamia ricini*, on day 7 just prior to hatching, acid phosphatase activity declined and alkaline phosphatase depicted a low activity till day 4 which however, got enhanced significantly on day 5 and remained almost constant till larva emergence. In *A. mylitta* both, acid and alkaline phosphatase activity increase with high peaks on days 3 and 6. High acid and alkaline phosphatase activity just before hatching of the first instar larva, has been noted in many lepidopterans[3,4,25].

Glycogen is one of the important stored metabolities. Its variation in *A. mylitta* (Fig. 3) reveals that with an initial decline on day 1 it increases till day 4 where after the concentration gets depleted very significantly by about 16·5% of the initial concentration. This decline could be attributed to its utilization for energy production. Contrary to this, Pant and Nautiyal[6], observed an increase thereof in *Philosamia ricini* during early developmental period where after with a slight decline till day 3 it maintained more or less a constant level till emergence.

A. mylitta during its embryogenesis exhibits a considerable variation in active phosphorylase activity (Fig. 3). It reveals a steady decrease on day 3 and then registers a sharp rise with the maximum activity on day 6 prior to hatching. Depletion of glycogen in the beginning and at the tail end of embryogenesis is in tune with the variation of active phosphorylase activity.

Total reducing sugars (Fig. 2) registered an initial concentration of 0·52 mg/100 mg dry weight and then declined at the time of hatching of the larva to 0·19 mg. Observations on several other insects[6,26,27], suggest the conversion of glycogen to reducing sugars. In *A. mylitta* however, although glycogen breakdown corresponds with active phosphorylase activity the variation pattern of the released reducing sugars does not appear to superimpose the glycogen breakdown pattern during embryonic development.

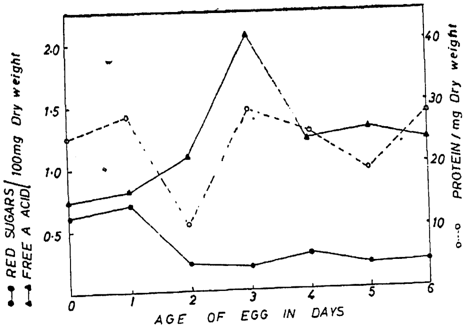

FIG. 2. Variation in proteins, reducing sugars and free amino acids during embryogenesis of *A. mylitta*.

The earlier view[28-31], that carbohydrates are utilized during early stages of development in insects has been confirmed by the present investigation on *A. mylitta*. With an initial decline till day 3, the total carbohydrates re-establish their original concentration on day 6 suggesting not only their initial utilization during early development but also the subsequent resynthesis thereof as development proceeds (Fig. 3).

During early stages of embryonic development, the pre-existing proteinous yolk reserves appear to undergo breakdown resulting in the lowering of protein concentration and a concurrent rise in that of free amino acids (Fig. 2).

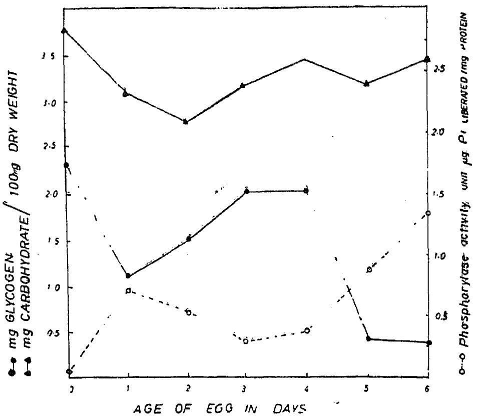

FIG. 3. Variation in glycogen, total carbohydrates and phosphorylase activity during embryogenesis of *A. mylitta*.

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1. Baker, R. J. and Alexander, B. H., *Ann. Ent. Soc. Amer.*, 1958, 51, 255.
2. Ashrafi, S. H. and Fisk, K. W., *Pakist. J. Sci. Ind. Res.*, 1961, 4, 70.
3. Sridhara, S. and Bhat, J. V., *J. Insect Physiol.*, 1963, 9, 693.
4. Pant, R. and Lacy, P. S., *Ind. J. Biochem.*, 1969, 6, 154.
5. — and Morris, I. D., *J. Biochem.* (Japan), 1969, 66, 29.
6. — and Nautiyal, G. C., *Proceedings. Ind. Acad. Sci.*, 1974, Sec. B, No. 3, 121.
7. Tichomiroff, A., Hoppe–Seyler's, *Z. Physiol. Chem.*, 1885, 9, 518.
8. Vaney, C. and Conte, A., *(Lyon.) Lab. d'Etudes de la Soie. Rpt.*, 1911, 14, 127.
9. Pigorini, L., *Russ. Ent. Obozr.* (Rev. Russed Ent.), 1925, 7, 181.
10. Crompton, M. and Birt, L. M., *J. Insect Physiol.*, 1967, 13, 1575.
11. Pant, R. and Sharma, S. C., *Ph.D. Thesis,* Allahabad University, 1967.
12. Colombo, G., Benassi, C. A. and Allegri, G., *Symposium on Germ Cells and Development International Inst. of Embryology and Fondazione,* A. Baselli, 1961, p. 354.
13. Indira, T., *Ph.D. Thesis,* Annamalai University, South India, 1963.
14. Shaw, E. I., *Expt. Cell. Res.*, 1955, 9, 489.
15. Bondansky, A., *J. Biol. Chem.*, 1933, 101, 93.
16. Sumner, J. B., *Science,* 1944, 100, 413.
17. Green, D. E. and Stumpf, P. K., *J. Biol. Chem.*, 1942, 142, 355.
18. Srivastava, L. M. and Krishnan, P. S., *Enzymologia,* 1961, 23, 270.
19. Wiens, A. W. and Gilbert, L. I., *J. Insect. Physiol.*, 1967, 13, 587.
20. Carrol, N. V., Longly, R. W. and Roe, J. H., *J. Biol. Chem.*, 1956, 220, 586.

21. Travelyan, W. F. and Harrison, J. S., 1952, 50, 298.
22. Nelson, D. H., *J. Biol. Chem.*, 1944, 153, 375; Somogyi, M., *Ibid.*, 1945, 160, 62.
23. Yemm, E. M. and Cocking, G. C., *Analyst*, 1955, 80, 209.
24. Lowry, O. H., Rosebrough, N. J., Farr, A. L. and Randall, R. J., *J. Biol. Chem.*, 1951, 193, 265.
25. Drilhon, A. and Busnel, R. G., *Bull. Soc. Chim. Biol.*, 1945 *a*, 27, 415.
26. Egorova, T. A. and Smolin, A. N., *Biokhimya*, 1962, 27, 476.
27. Dutrieu, J., *C.r. hebd. Seanc. Acad. Sci.*, Paris, 1961, 252, 347.
28. —, *C.r. Seanc. Soc. Biol.*, 1961, 253, 3071.
29. Urbani, E., *Acta Embryol et Morphol. Exptl.*, 1959, 2, 171.
30. — and Bellini, L., *Ricerca Sci.*, 1959, 29 (8), 1725.
31. Pant, R. and Lacy, P. S., *Ph.D. Thesis*, University of Allahabad, Allahabad, India, 1967, p. 101.

GEOCHEMICAL INDICATORS FOR GEOTHERMAL EXPLORATION

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Abstract

Of the various chemical parameters that can be applied to investigate potential geothermal areas, the most important are:

(*i*) elements associated with some late phase of magmatic or volcanic activity, *e.g.*, cesium.
(*ii*) Atomic ratio Na/K and the Na-K-Ca relationship.
(*iii*) Silica content of thermal spring water, and
(*iv*) the absence or near absence of some elements, *e.g.*, Mg, which are affected by temperature dependent reactions.

The appllication of these parameters in the case of thermal spring water from Puga (Ladakh) area, shows that the underground reservoir temperature, feeding the thermal springs, are likely to be around 200° C or even a little higher, indicating the strong potential of this area for exploitation of geothermal energy.

Introduction

Geochemical methods are now being increasingly employed to evaluate the various physicochemical parameters, which are important in the planning and operation of geothermal development programme. It has been shown that the silica content and the atomic ratio Na/K of the thermal spring water can be used to calculate reservoir temperatures (Fournier and Rowe, 1966; Mahon, 1966, White, 1965, 1967; Ellis, 1970). Recently Fournier and Truesdell (1972) have advocated the use of an empirical Na-K-Ca geothermometer for computation of reservoir temperatures. The use of other temperature dependent equilibria (*e.g.*, CO_2, H_2, CH_4, H_2O) in the gases discharged from the thermal springs and the associated carbon and sulphur isotopic ratios may also be used for estimating underground reservoir temperatures (Mahon, 1970).

Qualitative appraisal of reservoir temperatures can be made on the basis of presence or virtual absence of some elements like As, Hg, F, B, Cs, Mg, etc.

Gupta, M. L. and Sukhija (1974) and Gupta, Rao and Narain (1974) have recently applied the silica and the Na/K atomic ratio techniques to evaluate underground temperatures in Puga area. In the present paper apart from these two parameters, the Na-K-Ca method has also been used along with some qualitative parameters to estimate underground reservoir temperatures in Puga area.

Description of the Area

The Puga valley is located in the Ladakh District (Kashmir) and falls in the Survey of India Toposheet No. 52 K/8. It is situated at an altitude of 4400 m above m.s.l. and lies between the Zanskar and Ladakh ranges. The east-west trending Puga valley is about 15 km long and 1 km wide at its maximum and is surrounded by hills rising to a maximum elevation of 6033 m above m.s.l.

The climate is of the dry Arctic type, *i.e.*, hot days with very cold nights. Rainfall is scanty, but the higher peaks have some snowfall.

Geology

The country rock is quartz-mica-gneiss and quartz-mica-schist the latter often containing garnet (Sinha, 1971). The country rock contains micaceous quartzite sands and has been intruded by basic rocks occurring as sills and dykes. It is also traversed by a number of quartz veins.

The valley portion is covered by scree morraine and glaciofluvial and aeolian sands, which in turn are superficially encrusted upon by borax and sulphur. Borax surface encrustations occur in the central and eastern parts of the valley, the central portion having richer deposits than the eastern portion. Sulphur occurs as encrustations along fractures, joints and foliation planes in the quartz-sericite schists (which are locally gypsiferous). At the foot of the northern mountain, there is a very conspicuous thick sulphur and borax deposit, which at places is associated with gypsum, the latter apparently having been formed by the oxidation of sulphur.

The thermal waters issue out from over 100 springs, distributed all along the valley. In addition, 40–60 minor hot water seepages are also noticed. The majority of the hot springs fall in the temperature range 50° C to 70° C. The discharges of the thermal springs vary from a mere trickle to about 5 litres/sec. Some springs emanate small amounts of H_2S, CO_2, etc.

EXPERIMENTAL

About two and a half litres of water samples collected in polyethylene bottles were analysed for pH, HCO_3, CO_3, B, F, Cl, SO_4, NO_3, Ca, Mg, Na, K; sp. conductivity, SiO_2, total dissolved solids Fe, Mn, Br, I, Li, Rb and Cs by standard methods described in literature (Handa, 1974). Due to the possible presence of polymeric forms of silica, the samples were pre-treated with sodium hydroxide before applying the molybdate method.

RESULTS

While the actual chemical composition of the various thermal spring waters from the Puga area varies considerably, the salient features may be judged from the following data:

T° C	80		Cl	390	mg/l
pH	7·3		HCO_3	887	,,
Cond.	2690	micro-mhos/cm	SO_4	390	,,
			NO_3	tr	,,
TDS	2154	mg/l	B	116	,,
SiO_2	251	,,	F	12	,,
Ca	·8	,,	Fe	1·6	,,
Mg	2·7	,,	Br	1·2	,,
Na	596	,,	I	4·4	,,
Li	5·8	,,	Cs	11·3	,,
K	80	,,	Rb	·9	,,

Atomic Ratio

Na/K	12·65
Na/Rb	2593
Na/Li	29·8
Cl/B	1·05

DISCUSSION OF RESULTS

(A) *Qualitative Indicators*:

(*i*) *High cesium content.*—Because of its large ionic radius (1·67 Å), Cs^+ has a tendency to concentrate in the volatile phase during magmatic crystallization and may be strongly enriched in some pegmatites and pegmatite minerals, where its concentrations may even be high enough to form the mineral pollucite (Cs, Na) $AlSi_2O_6H_2O_1$. The relatively high cesium content of Puga thermal spring water (11·3 mg/1) seems to indicate its association with some late phase of magmatic or volcanic activity.

(*ii*) *Low magnesium content.*—The interaction of magnesium with clay is a temperature dependent reaction and magnesium is preferentially incorporated in the clay minerals which are stable at high temperatures. Consequently waters which have been in contact with rock minerals at 200°C (or more) should normally be highly depleted in magnesium, as is found to be the case with the Puga thermal spring water.

(B) *Quantitative Indicators*

(*i*) *The silica geothermometer.*—Assuming that at depth, the silica content in water is governed by the solubilty of quartz, Fournier (1970) has given two curves (Fig. 1) for estimating underground reservoir temperatures, depending upon whether the upward moving thermal spring waters are cooled entirely by adiabatic expansion at constant enthalpy (*i.e.*, steam losses occur due to decrease in hydrostatic pressure head) or whether heat losses occur by conduction into the wall rocks. The first curve is used for vigorously boiling springs with discharge rates in excess of 15–20 litres/sec., while the second curve is to be used for thermal spring having low rates of discharge (1–2 lit./sec.) and/or at below boiling temperatures.

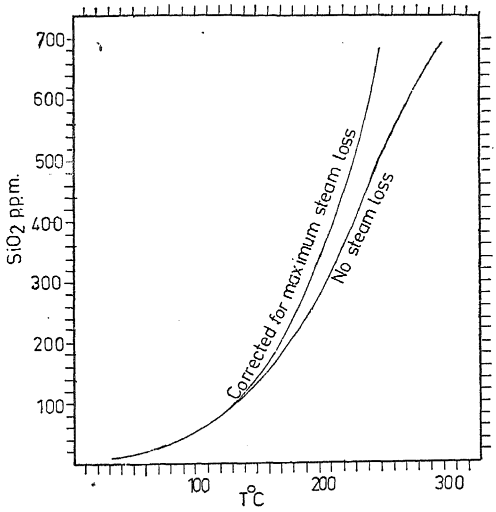

FIG. 1. Curves for estimating underground temperatures from the silica contents of waters discharged from hot springs.

Since the silica content of Puga thermal spring water is 251 mg/1, the use of these curves gives 183° C to 194° C, as the temperature range for the underground reservoir. This temperature range is likely to be the minimum, as some silica may be precipitated enroute and/or the silica content may be lowered through admixture with shallow aquifer waters.

(*ii*) *The Na/K geothermometer.*—The equilibrium constant K for the reaction: $K^+ + Na\text{-feldspar} \rightleftharpoons K\text{-feldspar} + Na^+$ is given by the equation:

$$K = \frac{(\text{K-feldspar})\,(\text{Na}^+)}{(\text{Na-feldspar})\,(\text{K}^+)} = \text{molar Na}^+/\text{K}^+$$

neglecting the effect of pressure and assuming that the activities of the solid species are unity and that the activity coefficients of Na^+ and K^+ cancel out each other, the variation of the equilibrium constant with temperature is given by the Van's Hoff equation, *viz.*,

$$\frac{d \log K}{d\,(1/T)} = -\,\frac{\Delta H^\circ\,(T)}{4 \cdot 578}$$

where T is the absolute temperature, ΔH° (T) is the standard heat of reaction at a given temperature. Since ΔH° (T) varies only slightly with temperature, a plot of log K, *i.e.*, log Na^+/K^+ (molar) *vs* 1/T can be used to determine the underground temperature. Ellis (1970) has drawn a curve (Fig. 2) showing the relationship between reservoir temperature and Na/K molar ratio. Since the Na/K molar ratio for Puga thermal spring water has a value of about 12·6, a reservoir temperature of 220° C is indicated. Recently Helgeson (1969) has given equilibrium constant values for the exchange reaction involving K-feldspar/albite and the use of these values gives a reservoir temperature of 204° C for the Puga thermal spring area.

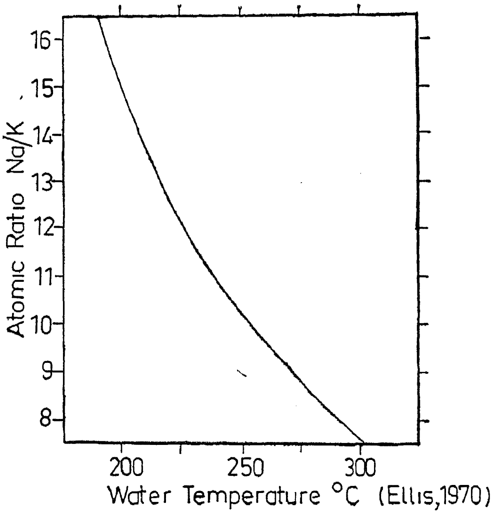

FIG. 2. The variation with temperature of the atomic ratio Na/K in waters in equilibrium with albite and feldspar.

(*iii*) *Na-K-Ca geothermometer*.—Since the exchange reaction between K-feldspar/albite may be affected by other cations, Fournier and Truesdell (1972) give the following equation for equlibrium constant involving Na, K and Ca, *viz.*,

$$\log K_c = \log \frac{Na^+}{K^+} + \beta \log \frac{\sqrt{Ca^{++}}}{Na^+}$$

$$= \frac{1647}{T^\circ (K)} - 2 \cdot 24$$

where T°(K) is the absolute temperature and β depends upon the stoichiometry of the reaction. For reactions above 100° C, a value of 1/3 for β has been suggested, while for reactions below 100° C a value of 4/3 for β has been empirically deduced by Fournier and Truesdell (1972).

From Fig. 3 and assuming a value of 1/3 for β, the reservoir temperature for Puga area comes out to be 240° C.

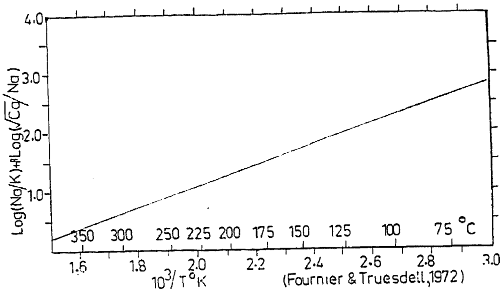

FIG. 3. Curves for geothermometry of thermal spring waters based on the Na-K-Ca relationships.

CONCLUSIONS

All the three quantitative geothermal indicators, *viz.*, the silica geothermometer the Na/K atomic ratio and the Na-K-Ca geothermal indicators give temperatures around 200° C for the underground reservoir in Puga area. The low content of Mg and relatively high cesium content also qualitatively support the above findings, indicating high potential of this area for geothermal exploitation.

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1. Ellis, A. J., *U.N. Symp. Development and Utilization of Geothermal Resources,* Pisa, 1970, p. 516.
2. Fournier, R. O. and Rowe, J. J., *Amer. Jour. Sci.,* 1966, **264**, 685.
3. —, *Symposium Hydrogeochemistry and Biogeochemistry,* Tokyo, Japan, 1970, p. 122.
4. — and Truesdell, A. H., *Geochim. et Cosmochim. Acta,* 1972, **37**, 1255.
5. Gupta, M. L. and Sukhija, B. S., *Geothermics,* 1974, **3** (3), 105.
6. —, Rao, G. V. and Narain Hari, *Geophys. Res. Bull.,* 1974, **12**, 119.
7. Handa, B. K., "Methods of collection and analysis of water samples and interpretation of water analysis data," *Tech. Manual, No. 1, Min. of Agr.,* 1974, p. 362.
8. Helgeson, H. C., *Amer. Jour. Sci.,* 1969, **267**, 729.
9. Mahon, W. A. J., *New Zealand J. Sci.,* 1966, **9**, 135.
10. —, *U.N. Symp. Development and Utilization of Geothermal Resources,* Pisa, 1970, p. 1310.
11. Sinha, B. P. C., *Proc. 58th Ind. Sci. Cong Assoc.,* Part III, 1971.
12. White, D. E., *Symp. Amer. Assoc. Petrol. Geol. Mem.,* 1965, p. 342.
13. —, *Amer. Jour. Sci.,* 1967, **265**, 641.

LETTERS TO THE EDITOR

PHOTON YIELD AND ENERGY IN INTERNAL BREMSSTRAHLUNG

ALTHOUGH a large number of measurements of IB spectra of both allowed and forbidden beta emitters have been carried out during the last few decades, most of the measurements either covered small energy ranges of spectra or made a relative comparison between experiment and theory for determining the contributions of various factors incorporated into the theory. While the earlier measurements[1-2], showed good agreement with Knipp and Uhlenbeck[3] and Bloch[4] (KUB) theory, later measurements[5-6], showed marked deviations especially at higher IB photon energies. Part of these discrepancies could be explained by incorporating Coulomb field effects[7-8], into the simple KUB theory and also by eliminating the contributions of EB to IB measurements. Latest measurements[9-10] have, however, pointed to the need of more accurate calculations.

Madansky and Rasetti[1], have reported the values of IB energy for ^{32}P and RaE which are 35% higher than the KUB theory. Similarly the results of Starfelt and Svantesson[11] for ^{35}S are higher than the KUB theory by 35%. Later measurements of Narasimhamurty and Jnanananda[5-6] from a number of allowed and forbidden beta emitters give values of IB energy which are higher by 25–100% than the Coulomb corrected theoretical values[7-8]. In the present communication we report our measurements of photon yield and IB energy for ^{32}P, ^{35}S, ^{45}Ca, ^{90}Y and ^{204}Tl and compare these with the various theories.

Beta radio-isotopes ^{32}P, ^{35}S, ^{45}Ca, ^{90}Y and ^{204}Tl were obtained from Bhabha Atomic Research Centre, Bombay, as H_3PO_4, H_2SO_4, $CaCl_2$, Sr $(NO_3)_2$ and Tl_2SO_4 in dil. HCl. Thin sources ($< 0\cdot1$ mg/cm^2) were prepared on a polythene film ($1\cdot47$ mg/cm^2) by evaporation method. The dimensions of the sources were 3 mm in diameter and their uniformity was obtained by addition of plain insulin solution. The IB spectra were recorded with a multichannel gamma ray spectrometer using NaI(Tl) detector in a geometry described elsewhere[9]. Absorbers of perspex were used to stop beta particles from reaching the detector. The IB spectra were corrected for room background, detector resolution, compton continuum, backscattered radiation, iodine escape peak, absorption in air, perspex absorbers, and detector covering. The contributions of EB were determined experimentally and subtracted from IB spectra. Using known values of detector efficiency and measured values of beta source strengths, values of photon yield and IB energy were determined by graphical integration. Details of the corrections and procedures for applying these are described elsewhere[9]. Experimental values of photon yield and IB energy (mc^2) per beta disintegration and the range of measurements are shown in Table I. The errors in the measurements, which include uncertainties due to various corrections, detector efficiency, statistical fluctuations and source strength measurements, are indicated along with the experimental values. Theoretical values calculated from various theories are also shown for comparison with the experimental results.

A comparison of experimental and theoretical values of photon yield and IB energy (mc^3) per beta disintegration shows that in the case of allowed beta emitters ^{32}P, ^{35}S and ^{45}Ca, the agreement with Nilsson KUB theory[7] is closest in the case of ^{32}P where the value of E_β^{max} is highest (1708 keV) but this agreement breaks down as the value of E_β^{max} decreases in the case of ^{45}Ca (258 keV) and ^{35}S (167 keV) with an increase in Coulomb effects on the emitted IB. The simple KUB[3-4] and Lewis and Ford[8], distributions give values which are still lower than the Nilsson KUB values[7]. In the case of forbidden beta emitters ^{90}Y and ^{204}Tl again none of the theories agrees with the experiment which gives substantially higher values. Thus we may conclude that the experimental values of

TABLE I

Photon yield and IB energy per beta disintegration

Betra emitte	E_{β}^{max} (keV)	Nature	Investigated range (keV)	IB photons per beta ($\times 10^4$)						IB Energy (mc^2) per beta ($\times 10^4$)					
				Experiment	Theory (1)	(2)	(3)	(4)	(5)	Experiment	Theory)1)	(2)	(3)	(4)	(5)
^{32}P	1708	allowed 1^+–0^+	50–1600	83±8	78·96	..	79·59	125·7	..	26·5±2·6	24·27	..	25·06	..	..
^{45}Ca	258	allowed $7/2^-$–$7/2^-$	30–240	4·0±0·4	3·490	3·696	3·715	..	..	0·33±0·03	0·272	0·296	0·313	..	..
^{35}S	167	allowed $3/2^+$–$3/2^+$	25–150	2·8±0·3	1·599	1·738	1·823	3·895	2·169	0·14±0·02	0·073	0·079	0·087	0·183	0·108
^{90}Y	2270	1st forbidden unique 2^-–0^+	550-1990	15·6±1·5	8·183	8·530	..	16·028	..	24· 4±2·4	12·646	13·602	..	25·80	..
^{204}Tl	766	1st forbidden unique 2^-–0^+	75–750	25·0±2·5	9·760	10·723	11·276	..	..	6· 1±0·6	2·654	3·010	3·336	..	..

Theory: (1) KUB theory; (2) Lewis and Ford KUB theory; (3) Nilsson KUB theory; (4) Felsner theory[12]; (5) Struzynski and Pollock[13] theory for ^{35}S.

photon yield and IB energy are consistently higher than the Coulomb corrected KUB values. Experimental IB intensity values are used in the calculations to take into account the Coulomb field and beta shape factor effects.

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1. Madansky, L. and Rasetti, F., *Phys. Rev.*, 1951, **83**, 187; and other references given there.
2. Boehm, F. and Wu, C. S., *Ibid.*, 1954, **93**, 518; and other references given there.
3. Knipp, J. K. and Uhlenbeck, G. E., *Physica*, 1936, **3**, 425.
4. Bloch, F., *Phys. Rev.*, 1936, **50**, 272.
5. Narasimhamurty, K. and Jnanananda, S., *Nuovo Cimento*, 1966, **46**, 217; and other references given there.
6. —. and —, *Proc. Phys. Soc.*, 1967, **90**, 109, and other references given there.
7. Nilsson, S. B., *Ark. Fys.*, 1956, **10**, 467.
8. Lewis, R. R. and Ford, G. W., *Phys. Rev.*, 1957, **107**, 756.
9. Powar, M. S. and Singh, M., *J. Phys.*, 1972, **5 A**, 460.
10. — and —, *Ibid.*, 1975, **1 G**, 453.
11. Starfelt, N. and Svantesson, N. L., *Phys. Rev.*, 1955, **97**, 708.
12. Felsner, G., *Z. Phys.*, 1963, **174**, 43.
13. Struzynski, R. E. and Pollock, F., *Nucl. Phys.*, 1966, **79**, 113.

A.C. VOLTAMMETRY AT THE TUBULAR GRAPHITE ELECTRODE

Most of the work in A.C. Voltammetry has been done on the cathodic side using the dropping mercury electrode. Only a few applications of the method using solid micro-electrodes have been reported in literature so far[1–5].

In order to carry out a.c. voltammetric measurements on the anodic side, involving organic compounds, it was considered advantageous to operate on a tubular graphite electrode[5] which offers stable and reproducible surface. Sargent Polarograph XXI was used in conjunction with a multi-frequency (1–5000 c.p.s.) plug-in auxiliary unit[7] in the present investigations. At the conventional frequency of 50 c.p.s., using o-dianisidine as the working material (10^{-4} M in 0·5 M H_2SO_4, flow rate 10 ml/min) the response was extremely poor. A high charging current with only a small kink at the halfwave potential was obtained. The base current after the kink, was found to be very much smaller than that before the kink.

Since the charging current varies directly with the frequency and the faradaic current with its square root, a better response of the electrode at lower frequencies could be expected. Experiments were, therefore, conducted at low frequencies in the range of about 1–20 c.p.s. using fresh electrode of exactly the same parameters, each time. Excellent a.c. peaks with good reproducibility were obtained only at low frequencies around 1 c.p.s. (Fig. 1). At such low frequencies, the recorder could easily follow the a.c. oscillations so that the voltammograms consisted of oscillating lines rather than smooth lines. The currents were, therefore, computed with respect to the mean positions of these oscillations. Good curves are obtained only at low frequencies around 1 c.p.s. With increasing frequencies, the curves become more and more unsymmetrical and are accompanied by an unproportionate increase in the charging currents (Fig. 1). At 50 c.p.s., the faradaic current was almost completely masked; only a small kink was noticed with a large difference in the base currents before and after the kink.

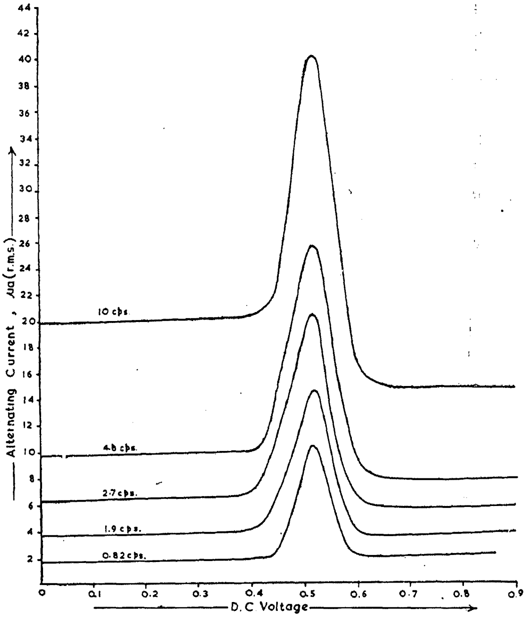

Fig. 1. A.C. Voltammograms obtained at different frequencies (using fresh electrode in each case) for the oxidation of *o*-dianisidine in 0·5 M H_2SO_4. Conc. $= 10^{-4}$ M, $V_f = 10$ ml/min., Amplitude of alternating voltage = 20 mV.

After ascertaining the frequency at which the tubular graphite electrode responded, the a.c. voltammograms were run for the other aromatic amino and phenolic compounds (anisidines, phenetidines, catechol, hydroquinone). The response was good.

According to Walker, Adams and Juliard[3], the a.c. peak current is almost independent of the hydrodynamics of the solutions. According to our observations, it is only in the case of o-dianisidine that the peak current is independent of the flow rate of the solution. In the case of other substances, the peak currents in stationary solutions are considerably smaller than those in moving solutions. Of course the value of the peak current is practically the same whether the flow rate is 1 ml or 20 ml per minute.

Walker *et al.*[3] also reported a.c. peaks for the oxidation of organic substances in aqueous as well as non-aqueous media, at platinum, carbon paste and graphite electrodes, at a frequency of 60 c.p.s. However, no such response was observed in our work. Even at the low frequencies it was only the tubular graphite electrode that gave a reproducible response. The response of the plain graphite electrode was only occasional and rarely reproducible.

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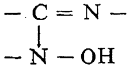
1. Juliard, A. L., Delaware Valley Conference, *Philadelphia, Pa.*, American Chemical Society (1957).
2. Juliard, A. L., *Nature*, 1959, **183**, 1940.
3. Walker, D. E., Adams, R. N. and Juliard, A. L., *Anal. Chem.*, 1960, **32**, 1526.
4. Vydra, F., Stulikova, M. and Petak, P. J., *Electronal Chem.*, 1972, **40**, 99.
5. Roizenblat, E. M., Levchenko, L. F. and Veretina G. N., *Zh. Anal. Khim.*, 1973, **28** (1), 33–41.
6. Sharma, L. R. and Dutt., J., *Ind. J. Chem.* (1968), **6**, 593, 597; (1969), **7**, 1220; (1970), **8**, 170.
7. Sharma, L. R., Dutt, J. and Kalia, R. K., *Ind. J. Tcch.* (in Press).

SPECTROPHOTOMETRIC DETERMINATION OF IRON(III) WITH N-HYDROXY-N-*p*-CHLOROPHENYL-N′-PHENYLBENZAMIDINE

ANALYTICAL potentialities of hydroxyamidines possessing bidentate functional grouping (I) have not been paid much attention. Only recently N-hydroxy-N, N′-diphenylbenzamidine has been applied for the analysis of metal ions[1,2]. The present paper reports the application of N-hydroxy-N-*p*-chlorophenyl-N′-phenylbenzamidine in the spectrophotometric determination of microgram quantities of Fe(III).



$$\begin{array}{c} -\mathrm{C}=\mathrm{N}- \\ | \\ -\mathrm{N}-\mathrm{OH} \end{array}$$

I

N-Phenylbenzimidoyl chloride obtained by Wallach method[3,4], by the action of thionyl chloride on benzanilide, was condensed with equimolar quantity of N-*p*-chlorophenylhydroxylamine in absolute ether. The hydrochloride thus obtained was converted into free base by ammonia and crystallised from petroleum ether (60–80°) : benzene (1 : 2). The pale yellow crystals of N-hydroxy-N-*p*-chlorophenyl-N′-phenylbenzamidine melted at 150° (Found : C, 70·95 ; H, 4·92 ; N, 8·45 ; $C_{19}H_{15}N_2OCl$ requires C, 70·72 ; H, 4·65 ; N, 8·67%).

With an ethanolic solution of the reagent iron(III) forms a blue coloured complex between pH 2·2–3·7. The colour is instantaneously produced and stable at least for 2 hrs. in the temperature range 15–40°. The visible spectra of the complex in aqueous ethanol shows a broad maxima at 580–600 nm. The wavelength, 590 nm (ϵ, 3685 ± 50 litre mole^{-1} cm^{-1}) was chosen for all absorbance measurements. The high sensitivity of the colour reaction permits the determination of iron at ppm level.

An ECI spectrophotometer model GS–865 and a systronic pH meter type 322 were employed for absorbance and pH measurements. Iron(III) stock solution was prepared by dissloving 1·0 g of pure iron wire (E. Merck) in 1 : 3 nitric acid and the solution boiled to expel oxides of nitrogen and diluted to a litre. Iron content of this solution was determined gravimetrically using 8-hydroxyquinoline[5].

To a suitable aliquot (2 ml) of solution containing 10–140 μg of Fe(III) and 2 ml of alcoholic reagent (0·1%) was added enough acetic acid or ammonium acetate so as to give a pH of 2·8 ± 0·1. Sufficient water and alcohol were then added to give a final volume of 10 ml of 60% ethanol (v/v) and allowed for 30 seconds and the absorbance at 590 nm was measured against alcohol as blank. The amount of iron is deduced from calibration curve.

The blue coloured complex obeys Beer's law over the range 10–140 μg (1·0–14·0 ppm) of the metal in a 10 ml solution at 590 nm (Table I). Practical range, as recommended by Sandell for this method, lies in the concentration 3·5–11 ppm of Fe(III).

Development of the colour commences at pH 2·2. Absorbance of the coloured complex is maximum and constant between pH 2·6–3·7. Accordingly a pH of 2·8 ± 0·1 was maintained for all analytical work. The absorption maximum of the complex shifts towards shorter wavelengths above pH 3·7.

Synthetic solutions containing a fixed amount of iron (5 ppm) and varying amounts of diverse ions were prepared. Measurement of colour developed in presence of alkali metals, alkaline earths, Pb(II), Zn(II), Cd(II), Hg(II), Bi(III); Cr(III), Mn(II), Ni(II), Co(II), chloride, sulphate and nitrate showed variations less than ± 0·02 units and did

TABLE I

Variation of absorbance of the blue complex with the concentration of iron(III)

Concentration of Iron (III) × 1·791 × 10^{-5} M	Absorbance
1	0·065
2	0·135
3	0·195
5	0·330
6	0·395
8	0·520
10	0.660
12	0·790
14	0·925

not interfere. However, Cu(II), Al(III), fluoride, tungstate, molybdate and citrate interfere seriously with the determination.

The mehod of Vosburgh and Cooper[6] showed the formation of only one complex under the conditions employed. Job's method of equimolar solutions and mole ratio method clearly indicated 1 : 3 stoichiometric ratio of the metal to ligand. Limiting logarithmic method[7] also confirmed this view. Thus N-hydroxy-N-*p*-chlorophenyl-N′-phenylbenzamidine affords as a sensitive method for the determination of iron(III) on a microscale.

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---

1. Satyanarayana, K. and Mishra Rajendra, K., *Anal. Chem.*, 1974, 46, 1609.
2. — and —, *Ind. Jour. Chem.*, 1975, 13, 295.
3. Wallach, O., *Ber. dt. chem. Ges.*, 1876, 9, 1212.
4. —, *Liebig's Ann.*, 1877, 184, 1 ; 1882, 214, 193; 226.
5. Vogel, A. I., *A Text Book of Quantitative Inorganic Analysis*, 3rd Edition, 1962, p. 524.
6. Vosburgh, W. C. and Cooper, G. R., *J. Amer. Chem. Soc.*, 1941, 63, 427.
7. Bent, H. E. and French, C. L., *Ibid.*, 1941, 63, 568.

SPECTROCHEMICAL PROPERTIES OF Cu(II), Ni(II) AND Co(II) COMPLEXES OF α-ALANINE

IN continuation to our previous work[1] on complexes of amino acids with transition metal ions, present communication describes the spectrochemical properties of the complexes of Cu(II), Ni(II) and Co(II) with α-alanine. The crystal field splitting parameter, $10D_q$, B and β have been calculated from the visible absorption spectra. The spectra have been recorded in the region 350–950 mμ, and indexed in Table I. The $10\,D_q$, B and β values are given in Table II.

TABLE I

Complexes of α-Alanine	λ_{max} in cm^{-1}	Transitions	∊
Cu(II)	15870	$^2E_g-{}^2T_{2g}$	28
Ni(II)	8700 (cal.)	$^3A_{2g}-{}^3T_{2g}$	
	15630 (obs.)	$-3T_{1g}$ (F)	23
	26310 (obs.)	$-{}^3T_{1g}$ (P)	
Co(II)	7474 (cal.)	$4T_{1g}-4T_{2g}$ (F)	
	16130 (cal.)	$-4A_{2g}$	25
	18860 (obs.)	$-4T_{1g}$ (P)	

TABLE II

Complexes of α-Alanine	$10\,D_q$ in cm^{-1}	B in cm^{-1}	β
Cu(II)	15870	..	..
Ni(II)	8700	1036	1·00
Co(II)	8600	831	0·85

Spectra of Ni(II) Complex

For divalent nickel, in octahedral coordination, three bands corresponding to the transitions $^3A_{2g}-{}^3T_{2g}$, $^3A_{2g}-{}^3T_{1g}$(F), $^3A_{2g}-{}^3T_{1g}$ should be observed. In the present investigations, however, only two bands are observed at 15630 cm^{-1} and 26310 cm^{-1} corresponding to the transitions $^3A_{2g}-{}^3T_{1g}$(F), and $^3A_{2g}-{}^3T_{1g}$ (P). Taking the band at 15630 cm^{-1} as ν_3 and using the equations of Ballhausen[2] the ν_1 band has been calculated. This gives the $10\,D_q$ value. The ratio ν_3/ν_1 comes out to be 1·80 indicating that the complex is octahedral[3-5]. Furthermore, the value for molar extinction coefficient is 25·0, confirming octahedral structure of Ni(II) alanine complex[6,7].

Spectra of Co(II) Complex :

Octahedral Co(II) complexes generally exhibit three bands around 460, 550 and 1150 cm^{-1} [8] associated with transitions $^4T_{1g}-{}^4T_{2g}$ (F), $^4T_{1g}-{}^4A_{2g}$, $^4T_{1g}-{}^4T_{1g}$(P). In the present case, however, only one band is observed around 18860 cm^{-1}. This can be attributed to ν_3 band. The ν_2 band was missing, may be due to the weak character of ${}^4T_{1g}$—${}^4A_{2g}$ transition. The values of 10 D_q, B and β have been calculated by employing the method described by Lever[9,10]. These values have been recorded in Table II and are indicative of octahedral Co(II) alanine complex[11].

Spectra of Cu (II) Complex

The absorption spectra of Cu(II) complex, in the present case, provides an asymmetric band around 15870 cm^{-1}. It is reasonable, therefore, to assign a distorted octahedral structure (12) to the system.

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---

1. Vasant Bhagwat, Vandana Sharma, Khadikar, P. V. and Kekre, M. G., *J. Inorg. Nucl. Chem.*, 1974, 36, 942.
2. Ballhausen, C. J., *Dan. Mat. Fys. Medel.*, 1954, 4, 29.
3. Manch, W. and Fernelis, W. C., *J. Chem. Educ.*, 1961, 38, 192.
4. Drago, R. S. and Rosenthal, *Inorg. Chem.*, 1965, 4, 840.
5. Gopal, Narayan, *Bull. Chem. Soc.* (Japan), 1966, 39, 2298.
6. Drago, R. S., *Inorg. Chem.*, 1963, 2, 124.
7. Chaston, *Aust. f. Shen.*, 1966, 19, 1041.
8. Costamagna and Levitus, R. J., *J. Inorg. Nucl. Chem.*, 1966, 10, 2685.
9. Lever, A. B. P., Lewis, J. and Nyholm, R. S., *J. Chem. Soc.*, 1964, p. 8761.
10. — and Nelson, S. M., *Chem. Comm.*, 1965, p. 168.
11. Lewis, J. and Wilkins, R. G., *Modern Coordination Chemistry*, Interscience, London, 1960.
12. Forester, D. and Goodgame, D. M. L., *J. Chem. Soc.*, 1964, p. 2790.

SPECTROPHOTOMETRIC ESTIMATION OF VANADIUM (V) AND TITANIUM (III) COMPLEXES WITH 4-METHYL DAPHNETIN

SPECTROPHOTOMETRIC and/or gravimetric estimation of Mo(IV), Nb(V), Ta(V), Th(IV), Fe(III) with 4-methyl daphnetin has been reported[1,2] however, there is no mention of V(V) and Ti(III) forming coloured complexes with this reagent. Our investigation relates to the use of 4-methyl daphnetin for the spectrophotometric determination of V(V) and Ti(III).

It is found that an alcoholic solution of 4-methyl daphnetin forms a green coloured complex with V(V) (λ_{max} 600 nm) and an orange coloured complex with Ti(III) (λ_{max} 410 nm) in the pH range of 5–7. Beer's law was verified upto 2 ppm in the case of V(V) and 8 ppm in the case of Ti(III). The molar-extinction coefficients are found to be $8 \cdot 5 \times 10^3$ and 8×10^3 for V(V) and Ti(III) respectively.

Estimation of Vanadium (V)

To a solution containing upto 2 μg of V(V) (aqueous NH_4VO_3) was added 2 ml of 0·1 M solution of 4-methyl daphnetin[3] and the resulting solution made upto 50 ml with distilled water. The colour development is almost instantaneous. The extinction of the complex was measured on spekol at 600 nm against the corresponding reagent blank. The amount of V(V) was then deduced from the calibration curve.

The effect of foreign ions was studied by adding to the V(V) solution (2 ppm) various cations and anions 10 times in excess. It was found that the ions, Co^{2+}, Ni^{2+}, Mg^{2+}, Sr^{2+}, Sn^{2+}, NO_3^{1-}, NO_2^{1-}, SO_4^{2-} Br^-, I^-, Cl^-, $[Fe(CN)_6]^{-4}$, CNS^-, did not interfere in the estimation; however, Fe^{2+}, Fe^{3+}, Ti^{3+}, Cu^{2+}, BrO_3^- were found to interfere.

Estimation of Titanium (III)

To a solution containing upto 8 μg of Ti(III) (alcoholic $TiCl_3$) was added 2 ml of 0·1 M solution of 4-methyl daphnetin and the resulting solution made upto 10 ml with ethanol. The experiment was carried out in alcoholic medium to keep the complex in solution. The extinction of the complex was measured at 410 nm with corresponding reagent blank. The amount of Ti(III) was then deduced from the calibration curve.

Ti(III) solution (2 ppm) was mixed with various cations and anions ten times in excess. It was found that Cu^{2+}, Co^{2+}, BrO_3^{1-}, CNS^- did not interfere in the estimation; however, Ni^{2+}, Fe^{2+}, Fe^{3+}, Mg^{2+}, NO_2^{1-} and $[Fe(CN)_6]^{4-}$ were found to interfere.

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---

1. Katyal, M., and Singh, H. B., *Talanta*, 1968, 15, 1043.
2. Patrovsky, V. (Central Geol. Inst. Prague, Chez), *Chem. Listy.*, 1961, 55 (8), 983 (*Analytical Abstract*, 1962, 2, 1325).
3. St. Von Kostaneki and Waber, C., *Ber.*, 1963, 26, 2906.

SYNTHESIS OF SOME SUBSTITUTED 4-THIAZOLIDINONE DERIVATIVES

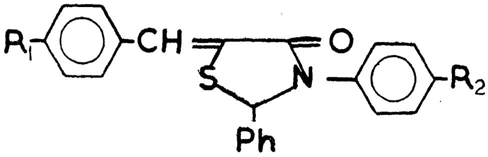
A RECENT publication by Harhash *et al.*[1], on the synthesis of 4-thiazolidinone derivatives prompts us to report some of our earlier work on these compounds which are known to exhibit interesting pharmacological properties[2].

The present work reports the synthesis of some new 2, 3-diaryl-5-arylidene-4-thiazolidinones obtained by the reaction of Schiff bases with β-aryl-α-mercaptoacrylic acids in boiling benzene solution. The thiazolidinones were colourless crystalline compounds (Table I) and were obtained in about 50–60% yields from A (acetic acid), B (benzene) or C (alcohol).

The u.v. spectra of these compounds showed λ_{max}^{MeOH} around 230–240 and 320–330 nm. The latter absorption is attributed to conjugation of the phenyl ring in the '5' position. The i.r., spectra (Nujol) showed bands at 1700 (C=O), 1500–1600 (aromatic) and 800–850 cm^{-1} (p-substituted phenyl ring).

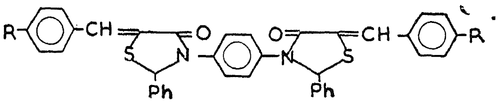
To confirm the 4-thiazolidinone structure of the compounds obtained by the above method, the compound VI was synthesised also by a different route. In this case, the Schiff base obtained by condensation of benzaldehyde with p-anisidine was reacted with thioglycolic acid as described by Surrey[3] to yield a thiazolidinone derivative which was condensed with p-chlorobenzaldehyde in presence of sodium ethoxide to afford VI obtained by the direct reaction of the appropriate Schiff base with β-p-chlorophenyl-α-mercaptoacrylic acid prepared as described in literature[4].

2, 3-Diaryl-5-arylidene- 4-thiazolidinones



No.	R_1	R_2	m.p. and Solvent	
I	Cl	COOH	280	(A)
II	Cl	Cl	185	(C)
III	Cl	CH_3	194	(B, C)
IV	Cl	Br	205	(B)
V	Cl	I	226	(B)
VI	Cl	OCH_3	198	(B, C)
VII	H	I	181	(C)
VIII	H	COOH	275	(A)

We also prepared thiazolidinone derivatives in about 40% yields by the condensation of the Schiff base, prepared from 2-moles of benzaldehyde and one mole of p-phenylenediamine with β-phenyl- and β-4-chlorophenyl-α-mercaptoacrylic acids in benzene solution to which the following structure was assigned. Both the compounds were obtained as white granular solids from acetic acid.



All the compounds gave satisfactory analysis, for C, H and N.

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1. Harhash, A. H., Elangdi, M. H. and Abdullah, S. O., *Indian J. Chem.*, 1973, **11**, 128.
2. Buu–Hoi, N., Saint–Ruf, G., Perche, J. C. and Bourglade, J. C., *Chem. Ther.*, 1968, **3** (2), 110; Surrey, A. R., *J. Am. Chem. Soc.*, 1949, **71**, 3354; Troutman, H. D. and Long, L. M., *Ibid.*, 1948, **70**, 3436; Surrey, A. R. and Culter, R. A., *Ibid.*, 1954, **76**, 578.
3. Surrey, A. R., *J. Am. Chem. Soc.*, 1947, **69**, 2911.
4. Campbell, N. and McKail, J. E., *J. Chem. Soc.*, 1948, p. 1251.

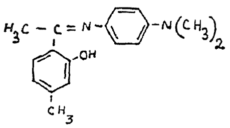
SOME TRANSITION METAL COMPLEXES WITH N-4-METHYL-2-HYDROXY ACETOPHENACYLIDENE-*p*-DIMETHYL AMINO ANILINE

CRYSTALFIELD splitting energy parameter Δ, Racah parameter B, Nephelauxetic coefficient β, magnetic moments and molar conductance have been determined for N-4-methyl-2-hydroxyacetophenacylidene-p-dimethylaminoaniline complexes with Cu(II), Ni(II) and Co(II). Spectrochemical series based on Δ was found in the order Cu(II) > Ni(II) > Co(II).

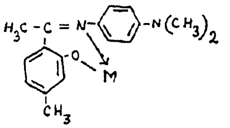
Experimental

The Schiff base N-4-methyl-2-hydroxyacetophenacylidene-p-dimethyl amino aniline was prepared by refluxing equimolar quantities of p-dimethylaminoaniline and 4-methyl-2-hydroxyacetophenone in ethanolic solution, on water-bath for 2·5 hrs. The contents were cooled, poured into water and the solid product obtained was washed with petroleum either (b.p. 60–80° C). Analytical data: (Found: C, 76·0; N, 10·23; H, 7·28; For $C_{17}H_{20}N_2O$

required: C, 76·12. N, 10·45; H, 7·46) support the preparation of the ligand (A) m.p. 178° C.



[A]



[B]

The complexes (B) of the metals Cu(II), Ni(II) and Co(II) were isolated[1,2], by refluxing stoichiometric mixture of the metal chlorides (A.R.) and the ligand in ethanol solution on water-bath for 1–2·5 hrs. The contents were cooled and the solid product was collected and crystallised from ethanol. Molar conductance of the complexes was determined in DMF solution and magnetic moment was measured in powder form at 30° C. Negative tests for metal ions in DMF solution indicate no dissociation of the complexes.

Results and Discussion

Ni(II) and Co(II) form 1 : 2 complexes while Cu(II) forms 1 : 1 complex. Copper(II) complex indicates its univalent nature[3] whereas Ni(II) and Co(II) complexes indicate unibivalent nature[3] in DMF solution. In the neutralisation of one gram mole of copper(II) complex only one gram equivalent of NaOH is required whereas for the remaining complexes two gram equivalents of caustic soda are required, showing thereby the monobasic and dibasic nature of the complexes respectively. Although magnetic data only cannot be used in deciding the geometry of complexes even then they are used to support it. Cu(II) having the electronic configuration $3d^9$ should exhibit a magnetic moment higher than that expected for one unpaired electron in tetrahedral complexes (1·75–2·21 B.M.). The paramagnetism observed for $H[Cu(C_{17}H_{19}N_2O)Cl_2]$ complex is consistent with the assignment of tetrahedral geometry[4]. Co(II) and Ni(II) having the electronic configuration, $3d^7$ and $3d^8$ should exhibit magnetic moments (4·0–5·3 B.M.) and (2·8–3·8 B.M.) respectively in octahedral complexes[5,6]. Further confirmation of geometry was made by molar extinction coefficient and by electronic spectral studies. Table I consists of electronic spectral, magnetic moment and molar conductance data of the complexes.

TABLE I

Complex	λ_{max} cm^{-1}	Molar extinction coefficient	Splitting energy Δcm^{-1}	Racah parameter B cm^{-1}	Nephelauxitic ratio β	λ_{max} Ohm^{-1} cm^2	Magnetic moment (B.M.)
$H[Cu(C_{17}H_{19}N_2O)Cl_2]$	22530	115	14,135	..	..	120	1·81
	14135	120					
$H_2 lNi(C_{17}H_{19}N_2O)_2Cl_2]$	28830	35	10,670	984	0·91	154	
	17945	40					2·98
	0670	38					
$H_2[Co(C_{17}H_{19}N_2O)_2Cl_2]$	25000	45	10,530	1097	0·98	158	
	20000	36					5·00
	9000	38					
$H_2[Co(C_{17}H_{19}N_2O_2)Br_2]$	21012	50	9,610	1001	0·89	170	
	17310	42					4·80
	8203	35					

B for Ni (II)and Co (II) ions was taken 1080 and 1120 respectively[10].

Spectral Studies

The spectrum of Cu(II) complex in DMF exhibits two absorption bands with peaks at 22530 cm^{-1} and 14135 cm^{-1}. The band at 22530 cm^{-1} is due to intraligand charge transfer[7,8] and the band at 14135 cm^{-1} is assigned to the transition $^2E_g \rightarrow {}^2T_{2g}$. Therefore, it seems to possess T_d configuration[4].

The spectrum of Ni(II) complex in acetone exhibits three absorption bands with peaks at 10670 cm^{-1}, 17945 cm^{-1} and 28830 cm^{-1}. These bands may be assigned to $^3A_{2g}$ (F) $\rightarrow$ $^3T_{2g}$ (F), $^3A_{2g}$ (F) $\rightarrow$ $^3T_{1g}$ (F) and $^3A_{2g}$ (F) $\rightarrow$ $^3T_{1g}$ (P) transitions respectively. This suggests octahedral coordination for Ni(II) complex in weak ligand field[9-12]. The ratio γ_2/γ_1 which lies between 1·6–1·8 further strengthens the coordination suggested above.

In the absorption spectrum of cobalt(II) complexes (in acetone), *viz.*, $H_2[Co(C_{17}H_{19}N_2O)_2Cl_2]$

and $H_2[Co(C_{17}H_{19}N_2O)_2\ Br_2]$ three bands with peaks at 9000 cm^{-1}, 8203 cm^{-1} (γ_1) ; 20000 cm^{-1}, 17,310 cm^{-1} (γ_2) and 25,000 cm^{-1}, 21,012 cm^{-1} (γ_3) were observed which are assigned to transition $^4T_{1g}(F) \rightarrow {}^4T_{2g}$ (F), $^4T_{1g}$ (F) $\rightarrow {}^4A_{2g}$ (F) and $^4T_{1g}$ (F) $\rightarrow {}^4T_{1g}$ (P). From these transitions octahedral coordination[10,12] for cobalt(II) complex is suggested. The ratio γ_2/γ_1 which lies between 1·90/2·4 also strengthens the above coordination[11,12].

Infrared Spectral Studies

In the i.r. spectra of metal chelates the stretching frequencies of azomethine $CH_3\ C = N$ (1600 cm^{-1}) and phenolic –OH (3400 cm^{-1}) groups are lowered and bands around 1490–1550 cm^{-1} and 3350–3400 cm^{-1} appear, showing thereby complex formation. This view is further supported by the formation of M–N (490–515 cm^{-1}) and M–O (460–478 cm^{-1}) bonds. In all these complexes metal to halogen bonds also formed as revealed by the appearance of bands around 350–375 cm^{-1}.

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1. Yamada, S., Kuge, Y. and Yamananouchi, K., *Bull. Chem. Soc.*, 1967, **40**, 1864.
2. —, — and —, *Co-ordin. Chem. Review*, 1966, **1**, 415.
3. Ghosh, S. P. and Bhattacharji, P., *J. Ind. Chem. Soc.*, 1974, **51**, 308.
4. Ozha, D. D. and Kaul, K. N., *Curr. Sci.*, 1974, **43**, 344.
5. Rastogi, D. K., Srivastava, A. K. and Jain, P. C., *J. Inorg. Nucl. Chem.*, 1970, **32**, 3949.
6. Carlin, R. L., *Transition Metal Chemistry*, Marcel Dekker, Inc., N.Y., Vol. 4, 1968.
7. Tanka, T., *J. Am. Chem. Soc.*, 1958, **80**, 4108.
8. Sheat, S. V. and Waters, T. N., *J. Inorg. Nucl. Chem.*, 1961, **19**, 73.
9. Ballhausen, C. J., *Introduction to Ligand Field Theory*, McGraw-Hill, Book Co., N.Y., 1962.
10. Figgis, B. N., *Introduction to Ligand Fields*, John Wiley and Sons. Inc., N.Y., 1967.
11. Holms, O. G. and Mcclure, D. S., *J. Chem. Phys.*, 1957, **26**, 1686.
12. Singh, P., Singh, V., Singh, B. P. and Mahesh, R. P., *Ind. J. Chem.*, 1975, **13**, 734.

EFFECTS OF RIFAMPICIN AND NITROFURANTOIN ON ^{14}C AMINO ACIDS INCORPORATION BY DIFFERENT SUBCELLURAR FRACTIONS FROM MOUSE SKELETAL MUSCLE AND FIBROSARCOMA

RIFAMPICIN AND NITROFURANTOIN are two potent inhibitors of macromolecule synthesis in different subcellular fractions. While the mode of action of nitrofurantoin is obscure, that of rifampicin is much debated. Rifamycins are known to inhibit bacterial[1], as well as nucleolar and nucleoplasmic[2] and mitochondrial[3] RNA polymerases. In addition rifamycins cause inhibition of polyphenylalanine synthesis in cell-free systems from bacteria[4] animal cells[2]. It has been reported from this laboratory that rifampicin inhibits polyphenylalanine synthesis by intact mitochondria and that this inhibition cannot be released by poly (U)[5]. Similar results have also been reported by others[6]. On the other hand it has also been reported that poly (U) can reverse the inhibition of polyphenylalanine synthesis in a cell-free system consisting of mitochondrial ribosomes and supernatant proteins[7]. The mode of action of nitrofurantoin has not been studied in detail. Röschenthaler *et al.*[8], suggested that the inhibition of growth and IPTG induced β-galactosidase synthesis in *E. coli* by nitrofurantoin could be due (*i*) to interference with energy metabolism or (*ii*) to a disturbance of the cell membrane or (*iii*) to a more direct action on the protein synthetic machinary. But we have previously reported that in mitochondria the inhibition of protein synthesis is not due to an interference with the energy metabolism[9].

Materials and Methods

L[U-^{14}C] lysine (specific radioactivity, 114 mCi/mmol) was obtained from Bhabha Atomic Research Centre, Bombay, India. Rifampicin was a gift from CIBA, Ltd., Basel, Switzerland and Cycloheximide, Chloramphenicol, Nitrofurantoin, poly (A), ATP, GTP, Phosphoenol pyruvate, Pyruvate kinase were purchased from Sigma Chemicals, Mo., U.S.A. All other chemicals were of analar grade.

Mitochondria were isolated according to the method of Chakrabarti *et al.*[10], and further purified by washing at 7000 g as described by O'Brien and Kalf[11]. Microsomes and pH 5 enzymes were prepared following the method of Dube *et al.*[12]. Mitochondria and microsomes were incubated in mediums identical to those described by Dube *et al.*[12]. The reaction was terminated by adding TCA containing sodium tungstate to a final concentration of 5% TCA and 0·25 *M* sodium tungstate and the radioactivity was determined as described elsewhere[12].

TABLE I

Effects of rifampicin, nitrofurantoin and poly (A) on the incorporation of ^{14}C-amino acid by different subcellular fractions from mouse skeletal muscle and fibrasarcoma

(Results are expressed as counts/min/mg. of protein and represents values for 6 different experiments with ± S.D.)

Incubation system	Mitochondria		Microsomes	
	Normal	Malignant	Normal	Malignant
Complete	1463±65	2172±100	2108±109	3843±215
„ − pH 5 enzymes	..	..	420± 22	1924± 71
„ + Cycloheximide (100 μg)	1297±59	2156±101	1024± 60	1565± 66
„ + Chloramphenicol (100 μg)	507±26	1060± 58	1911± 67	3904±200
„ + Poly (A) (100 μg)*	1970±73	3208±120	2810±119	4476±189
„ + Rif. 10 μg *	660±28	1040± 56	722± 32	1924± 69
„ + Poly (A) (100 μg) + Rif (10 μg) *	710±30	1654± 70	98± 36	3236±240
„ + Poly (A) (100 μg * + Rif (10 μg)	750±33	1908± 75	815± 40	3560±210
„ + NFT (20 μg) *	433±22	1117± 80	1052± 55	2304±100
, + Poly (A) (100 μg)* + NFT (20 μg)	620±27	1535± 65	890± 50	2850± 99
„ + Poly (A) (100 μg) + NFT (20 μg)*	706±30	1500± 63	923± 56	2696±105

* Different subcellular fractions were pre incubated with either poly (A) or rifampicin (Rif) or nitrofurantoin (NFT) for 15 min at 37° C.

From Table I it will appear that cycloheximide and chloramphenicol selectively inhibit microsomal and mitochondrial protein synthesis respectively. This is in accordance with previous findings[13,14]. With poly (A) there is significant increase of lysine incorporation into all subcellular fractions studied. Rifampicin inhibits lysine incorporation into all subcellular fractions, but a reversal of this inhibition by poly (A) to any significant extent is observed only in the case of mitochondria and microsomes isolated from malignant tissues. But a careful consideration of these and previously reported data[4-7] indicates that in either case rifampicin must affect protein synthesis at the level of translation.

Table I also shows that nitrofurantoin inhibits lysine incorporation by all subcellular fractions. Since we have already reported that nitrofurantoin induced inhibition is not due to an interference with energy metabolism[10] and since the question of disturbance of membrane does not arise in case of a cell-free system consisting of microsomes and pH 5 enzymes it appears that nitrofurantoin directly affects the protein synthesis apparatus at least in the microsomal system. The slight increase of incorporation when malignant mitochondria or microsomes are pre-incubated with either nitrofurantoin or poly (A) seems to be the effect of pre-incubation on the incorporation rather than an effect of poly (A).

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1. Wehrli, W. and Staehelin, M., *Bact. Rev.*, 1971; 35, 290.
2. Busiello, E., Girolamo, A. D., Girolamo, M. D.; Fantuzzi, L. F. and Vesco, C., *Eur. J. Biochem.*, 1973, 35, 251.

3. Wu, G. J. and Dawid, I. B., *J. Biol. Chem.*, 1974, 249, 4412.
4. Calvori, C., Frontali, L., Leoni; L. and Tecce, G., *Nature*, 1965, 207, 417.
5. Duhe, D. K., Chakrabarti, S., Sarkar, A., Bhattacharya, P., Goswami, B. B. and Roy, S. C., *Biochem. Pharmacol.*, 1973, 22, 659.
6. Grant, W. D. and Poulter, R. T. M., *J. Mol. Biol.*, 1973, 73, 439.
7. Goswami, B. B., Chakrabarti, S., Dube, D. K. and Roy, S. C., *Physiol. Plantarum*, 1974, 32, 291.
8. Röschenthaler, R., Kindler, P., Herrlich, P. and Igbokwe, J., *Zbl. Bakt. I. Abt. Orig.*, 1970, 215, 203.
9. Goswami, B. B., Chakrabarti, S., Bhattacharya, P., Sinha, S. N., Roy, B. K., Roychowdhuri, P. and Dube, D. K., *Biochemi. Pharmacol.*, 1974, 23, 470.
10. Chakrabarti, S., Dube, D. K. and Roy, S. C., *Biochem. J.*, 1972, 128, 461.
11. O'Brien, J. W. and Kalf, G., *J. Biol. Chem.*, 1967, 247, 2172.
12. Dube, D. K., Chakrabarti, S. and Roy, S. C., *Cancer*, 1972, 29, 1575.
13. Sisler, H. D. and Siegel, M. R., *Antibiotics*, 1967, 1, 283.
14. Ashwell, M. A. and Work, T. S., *Ann. Rev. Biochem.*, 1970, 39, 251.

ON THE SIGNIFICANCE OF CALCITE CEMENTATION IN GARUDAMANGALAM LIMESTONE, TRICHINOPOLY UPPER CRETACEOUS, SOUTH INDIA

GARUDAMANGALAM Limestone is a thin elongated, impersistent, shell bearing calcareous sandstone occurring at the base of Trichinopoly Group of Blanford (1855) or the Garudamangalam Group of Rama Rao (1956) in the exposed upper cretaceous sequence of South India. Petrographically, Garudamangalam Limestone shows a floating framework of elongated but subrounded quartz and feldspar grains, heavy minerals and shell fragments held in a medium of coarse, blocky, sparry calcite cement. Modally calcite cement constitutes about 48 to 58% of the rock. Much of quartz and feldspar grains are corroded by blocky calcite cement more along the longer sides of the grains than at their terminal ends. The form, nature, size and disposition of calcite cement suggest that it was deposited in the abundant pore space of the original quartz, feldspar, shell framework by slow percolating solution (Dapples, 1971).

Numerous field and petrographic evidences are presented by Rao and Siddiqui (1974) to show that Garudamangalam limestone is an ancient analog of a modern sandy beach rock. Folk (1974) has recently discussed the natural history of calcium carbonate and has shown that modern beach rocks represent high Mg^{++} and high Na^{+} environment and therefore beach rock cementation is either by micritic or fibrous aragonite, or micritic and bladed or fibrous high Mg–calcite. Low–Mg sparry calcite cement is rare in such an environment. If this is true, then Garudamangalam limestone shows sparry calcite cement and therefore may not be a beach rock, whereas other field and petrographic evidences reveal that Garudamangalam limestone is indeed a beach sediment. In order to clear the doubt whether the sparry calcite cement in Garudamangalam limestone is indeed a low-Mg calcite, X-ray diffractograms were taken from the cement portion of Garudamangalam limestone by a General Electric, XRD–6 X-ray diffractometer ($K\alpha$ radiation) with 2° 2θ rotation per minute over the range of 25° to 32°, 2θ angle (Fig. 1). Invariably it turned out to be low-Mg calcite (3·029 Å *d*); then here is a problem where an ancient beach rock shows a low-Mg, sparry calcite cement.

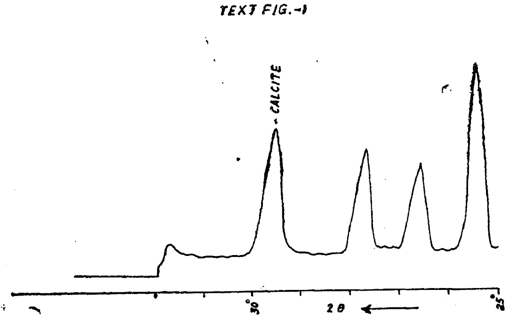

FIG. 1. X-ray diffractogram of sparry calcite in Garudamangalam Limestone, Trichinopoly cement Upper Cretaceous, S. India. Note the prominent peak for calcite corresponds to Mg-poor calcite.

Folk (1974) discusses this issue and suggests two alternatives for low-Mg calcite cementation in ancient beach rocks: (1) exposure to surface conditions where the beach comes into contact with the surface fresh waters low in Mg^{++} or (2) deep burial where the beach sediment comes into contact with Mg^{++} depleted connate or meteroic water diluted subsurface brines. The Mg^{++} is removed at the shallower levels, either by clay minerals or by capture by replacement dolomite. The option with reference to Garudamangalam limestone is for the second alternative for, the sparry calcite cement here is blocky, absence of solution deposition babrics in the associated shell debris and the absence of micritic or fibrous calcite crust to open spaces giving place centropetally to coarse mosaic spar.

There is also a suggestion (Folk *op. cit.*) that modern seas are rich in Mg^{++} and Na^{+} and poor in Ca^{++} as most of Ca^{++} is extracted by marine organisms to build their calcareous skeletons. Mg^{++} concentration in marine waters has progressively increased from Cretaceous times as the proliferation of calcareous marine organisms is unprecedented since cretaceous. Pre-cretaceous waters were relatively rich in Ca^{++} and therefore most of the carbonate cementation was by low-Mg calcite. If this is so, it is to be seen that the upper-cretaceous section of Trichinopoly area is prolific in calcareous organisms both invertebrates, benthonic and pellagic foraminifera. There must have meen enough Mg^{++} in the upper cretaceous marine waters of Trichinopoly Cretaceous to cause high Mg-calcite penecontemporaneous cementation in Garudamangalam limestone. The very fact, that it has a low Mg-sparry calcite cement suggests that carbonate cementation in this formation is post-burial, that is late diagenetic.

The time of cementation has a point in petroleum geology. If the cementation is early diagenetic in a type of rock that is under consideration, even though there may be oil in the basin, the reservoir rock, as for example a beach sediment remains dry. If the cementation is late diagenetic and migration of oil is early then the beach sediment would be a prolific producer. Per contra, in the subsurface a shore line rock as the sandy beach rock discussed here, is proved to carry late diagenetic cement, even though there may be oil in the basin it has not migrated early or as the chances, are, there is no easily migratable oil in the basin.

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1. Blanford, H. F., "On the cretaceous and other rocks of the South Arcot and Trichinopoly Districts," *Mem. Geol. Surv. of Ind.*, 1865, 4.
2. Dapples, E. C., "Physical classification of carbonate cement in quartzose sandstones," *Jour. Sed. Pet.*, 1971, 41, 196.
3. Folk, R. L., "The natural history of crystalline calcium carbonate. Effect of magnesium content and salinity," *Ibid.*, 1974, 44, 40.
4. Rao, Gundu C. and Siddiqui, Z. R., "Garudamangalam limestone, of the Trichinopoly Upper Cretaceous, South India," *O. N. G. C. Bull.*, 1974 (In press).
5. Rama Rao, L., "Recent contributions to our knowledge of the Cretaceous rocks of South India," *Proc. Ind. Acad. Sci.*, Spl. Publ., Vol. 44, 1956.

SUSCEPTIBILITY OF TWO NEOPLASTIC CELL LINES, *VIZ.*, MOUSE FIBROSARCOMA (MFS-8) AND HUMAN FIBROSARCOMA (HFS-9) TO INFECTION WITH ARBOVIRUSES ISOLATED IN INDIA

STUDIES on the arbovirus susceptibility of human synovial cell line (McCoy) was reported earlier[2]. The present communication reports the virus susceptibility of two more cell lines of neoplastic origin, *viz.*, mouse fibrosarcoma (MFS-8)[1,5], human fibrosarcoma (HFS-9)[5].

Mouse fibrosarcoma (MFS-8) cell line was developed by Ranadive *et al.*[1,5] and was maintained on basal medium Eagle (BME) with 20% horse serum with supplementation of cystine (15 mg/litre of BME). Further, both the cell lines were adapted "at VRC" on minimum essential medium (MEM) (Dulbecco) with Earles base and supplemented with 12% goat serum (GS). Cells were passed every 7th day and the medium was renewed on the 4th day. The split ratio was 1 : 12 and work was done between passage levels 25–39. For human fibrosarcoma (HFS-9), the split ratio was 1 : 15 and the work was done between passage levels 65–75.

The following arboviruses were employed in the present study:

Antigenic Group	Name of the virus, abbreviation, strain No. and passage history
A	: Sindbis (SIN) AR339 M_5V_2; Chikungunya (CHIK) (634029) M_{11} V_1; Semliki Forest Virus (SF)* (718488) $M_{10}V_1$.
B	: West Nile (WN)* (E101) M_5V_2; Japanese Encephalitis (JE) (P20778) M_9; Dengue–I (DEN–1)* (Hawaii) M_{88}; Dengue–II (DEN–2) (P23085) M_{41}; Dengue–III (DEN–3) (633798) M_{28}; Dengue–IV (DEN–4) (611319) M_{28}; Kyasanur Forest Disease (KFD) (P9605) M_{15}.
BUNYAMWERA	: Batai (BAT) (G20217) $M_{12}V_1$.
SIMBU	: Sathuperi (SAT) (G11155) M_{13}; Ingwavuma (ING) (633970); M_7BH_1; Thimiri (THI) (66414) M_{13}
VSV	: Chandipura (CHP) (653514) $M_{22}V_1$.
AHS	: African Horse Sickness (AHS) (603395) M_{102}.
TURLOCK	: Umbre (UMB) (G1424) M_8.
PALYAM	: Palyam (PAL) (G5287) M_{10}; Vellore (VEL) (68886) M_{12}; Kasba (KAS) (G15534) M_{21}.
SANDFLY FEVER	: Sandfly fever (SFS) (671417) (Sicilian) BS-C-1, BH_{12}.
GANJAM	: Ganjam (GAN) (G619) M_5BH_1.
KAISODI	: Kaisodi (KSO) (G14132) M_{43}.
KEMEROVO	: Wadmedani (WM) (G673) M_{12}.

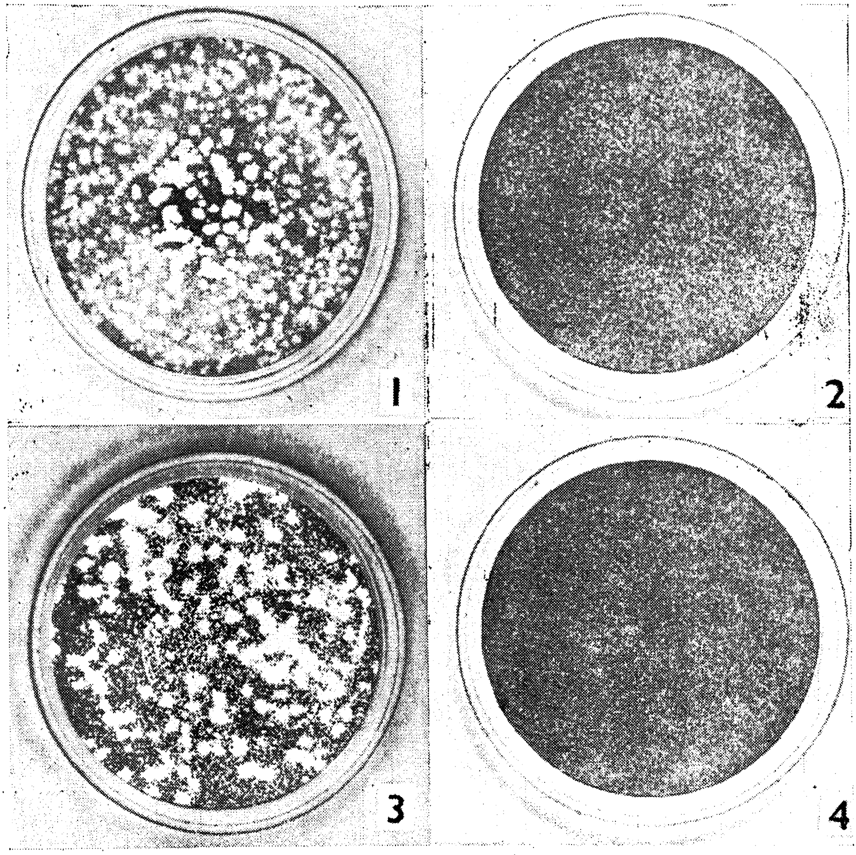
Antigenic Group		Name of the virus, abbreviation, strain No. and passage history
UNGROUPED	:	Arkonam (ARK) (G10658) M_{40}; Minnal (MIN) (G7481) M_{33}; Thottapalyam (TPM) (66412) M_{16}; Kannamangalam (KAN) (66415) M_{22}; Sembalam (SEM) (66416) M_{17}; Bhanja (BHA) (G690) M_5BH_1; Dhori (DHO) (611313) M_7; Wanowrie (WAN) (G700) M_{19}.

M — Suckling mouse brain passage; V — Vero passage; BH — Baby hamster kidney (BHK-21) passage; BSC–BS-C-1 passage;

* Not isolated at VRC.

Methods for preparation of virus stocks; inoculation, detection, gradation of cytopathic effect (CPE) and virus assay were followed as described earlier[2,3,6]. The infected tubes were harvested on the 10th post-inoculation day (PID) instead of 5th (PID) as described earlier[2]. The infected tissue culture fluids (ITCF) at third passage level were passaged in susceptible host systems *e.g.* Vero, BS-C-1 and BHK-21 cell lines or suckling mice. If there was CPE in cells or illness and death in mice the fluid was then titrated in the same host system. In plaque formation studies, the overlay contained 0·5% methyl cellulose (METHOCEL) instead of carboxymethyl cellulose (CMC).[4]

Out of the thirty-two viruses SIN, SF, CHIK, WN, JE and CHP, viruses multiplied in MFS-8 cell line and CHIK, SF, KFD and CHP in HFS-9 cell line; others did not multiply, nor showed CPE or plaque formation. SF and CHP viruses produced CPE and plaques in both the cell lines. CHP plaque size was 0·8 mm in MFS-8 (Fig. 1) cell line and 0·7 mm in HFS-9 (Fig. 3) cell line. Controls for MFS-8 and HFS-9 are shown in Figs 2 and 4. SF plaque size was 3·3 mm in MFS-8 cell line and 1 mm in HFS-9 cell line.



FIGS. 1–4. Fig. 1. Chandipura plaques in MFS-8 cells, × 3. Fig. 2. Control MFS-8 cells, × 3. Fig. 3. Chandipura plaques in HFS-9 cells, × 3. Fig. 4. Control HFS-9 cells, × 3.

MFS-8 and HFS-9 cell lines were found to be poorly sensitive to arboviruses with respect to other standard cell lines such as Vero, BHK-21, HeLa, HEp-2, etc., as determined by CPE multiplication and plaque formation studies.

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Virus Research Centre, S. S. GOGATE.
(Indian Council of Medical Research),
Poona 411001, *April* 7, 1975.

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1. Gangal, S. G., Bapat, C. V. and Ranadive, Kamal J., *Indian J. Path. Bact.*, 1959, **2**, 103.
2. Gogate, S. S., *Indian J. Med. Res.*, 1974, **62**, 684.
3. Haldane, J. B. S., *Nature*, 1960, **187**, 879.
4. De Madrid, A. T. and Porterfield, J. S.. *Bull. Wld. Hlth. Org.*, 1969, **40**, 113.
5. Ranadive, Kamal J., Bhisey, A. N. and Wagh, U. V., *Bull. Nat. Inst., Sci. India*, 1962, **19**, 259,
6. Reed, L. J. and Muench, H., *Am. J. Hyg.*, 1938, **27**, 493.

PANCREAS IN CERTAIN FRESH WATER TELEOSTS

THE occurrence and distribution of pancreatic tissue have been known to vary in fishes. In *Cyprinus carpio* (Curry, 1939) it is a diffuse mass lying in between the loops of the intestine. Sarbahi (1939) recorded the pancreas of *Labeo rohita* as masses of tissue all over the viscera and also within the substance of the liver. Lal *et al.* (1964) also observed penetration of the pancreas into the liver in *Chagunius chagunio*. In *Gadus morhua*, it has been found to be distributed in between the pyloric caeca along the bile and hepatic ducts Bishop and Odense, (1966). Kristal (1946) has observed two types of distribution, a diffuse type with the pancreas on the mesentery alone, without penetration into liver, and a dispersed type, with the pancreas penetrating into liver.

Three species of fishes were examined in the present study : *Glossogobius giuris* (Carnivorous), *Rasbora daniconius* (Omnivorous) and *Puntius conchonius* (Herbivorous). Both diffuse and dispersed types of pancreas (of Kristal's classification) were found in *Puntius conchonius* and *Rasbora daniconius* (Fig. 1). However, in *Glossogobius giuris*, the pancreas is a distinct compact gland. Also in this species, the exocrine pancreas is found to penetrate into the endocrine pancreas (Fig. 2). Such penetration has been reported in a few other teleosts also (McCormic, 1924).

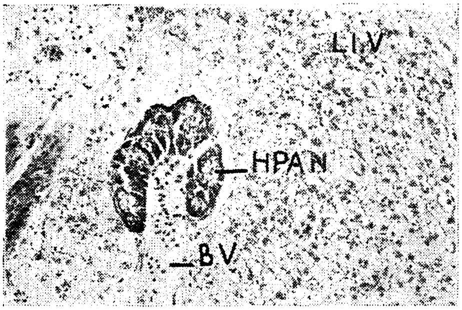

FIG. 1. Photograph showing penetration of exocrine pancreas into the liver : *Rasbora daniconius*, × 320.

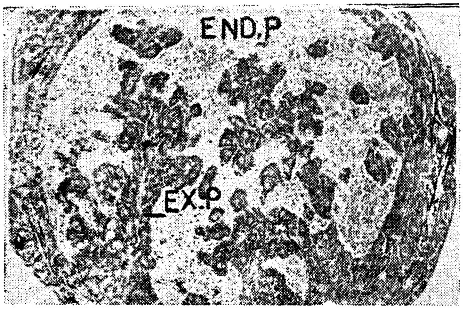

FIG. 2. Photomicrograph showing penetrating of exocrine pancreas into the substance of endocrine pancreas : *Glossogobius giuris*, × 80.

(BV, Blood Vessel ; END P, Endocrine Pancreas ; EX P, Exocrine Pancreas ; H PAN, Pancreas, LIV, Liver.)

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---

1. Bishop, C. and Odense, P. H., *J. Fish. Res. Bd. Canada*, 1966, 23, 1607.
2. Curry, E., *J. Morphol.*, 1939, **65**, 53.
3. Kristal, J., *S. Afr. J. Med. Sci.*, 1946, **11**, 79.
4. Lal, M. B., Bhatnagar, A. N. and Kaile, R. K., *Proc. Nat. Acad. Sci. India*, 1964, **34 B**, 160.
5. McCormick, N. A., *Trans. Roy. Canadian Inst.*, 1924, 15, 57.
6. Sarbahi, D. S., *J. Roy. Asiatic Soc. Bengal*, 1939, 5 (2), 87.

AN UNDESCRIBED SPECIES OF *MICROCLAVA* (HYPHOMYCETE) FROM KARNATAKA

AN interesting but a rare hyphomycetous fungus was collected recently from the forests of Agumbe (India) which showed the following characters:

Colonies effuse, non-conspicuous; mycelium superficial; conidiophores simple, unbranched, erect, slightly bulged at the apex into an obconical or ellipsoidal structure with two sporogenous cells; conidia (aleuriospores) 1-celled, thick-walled, faintly verrucose, brown, borne side by side in pairs, subglobose with flattened base; on the basis of which it was identified as a species of *Microclava* Stev.[1,2]. A thorough perusal of literature revealed no report of this genus from India so far[3]. The Indian collection of the fungus was compared with the other three known species[1] *viz.*, *Microclava miconiae* Stev., *M. bispora* (Hansf.) Deighton, and *Microraimensis* (Batista and Cavalcanti) Deighton, all reported from U.K. The present collection significantly varied from these species in respect of morphology. However it showed some resemblances with *M. bispora* but differed from it in possessing shorter conidiophores and larger conidia; and hence the same has been presented here as a new species:

Microclava indica sp. nov. (Fig. 1)

Coloniae effusae, non-conspiceosae et non-visiblis; mycelium superficiale; septatae; conidiophora simplicia, recta, septata, non-ramosa, macronematae et mononematibus, pallide-brunneae, magnit 28–40 × 3·5–4 μ; conidia (aleuriosporae) sub-spherica vel globosa, unicellularia, brunnea, crassiuscule tunicati, leviter verrucosa, 4–10 × 4–7 μ.

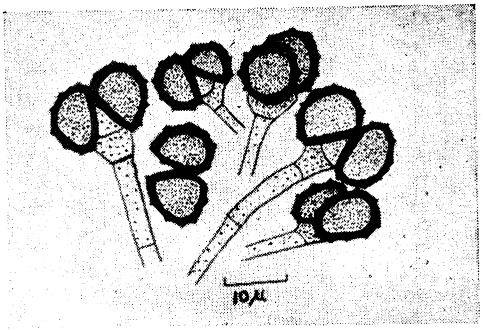

FIG. 1. Showing conidiophores and conidia of *Microclava indica* sp. nov.

Matrix.—On leaves of *Schleichera* sp. (Fam: Sapindaceae) legit: AWS (16th December 1974) at Agumbe, AMH No. 2533 (Holotype).

Remarks.—This is a new generic record to India and was found closely associated with a microthyriaceous fungus in nature.

Grateful thanks are due to Prof. M. N. Kamat for his keen interest and to the Ministry of Education, Govt. of India, for the award of S.R.T. to the senior author.

M.A.C.S. Research Institute, A. W. SUBHEDAR.
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April 28, 1975.

---

1. Deighton, F. C., *Microclava*, *Trans. Brit. Myc. Soc.*, 1969, **52**, 315.
2. Ellis, M. B., *Dematiaceous Hyphomycetes*, C.M.I., Kew, 1971, p. 109.
3. Mukerji, K. G. and Juneja, R. C., *Fungi of India*, Emkay Publ., Delhi, 1975, p. 224.
4. Subramanian, C. V., *Hyphomycetes*, I.C.A.R. Publ., New Delhi, 1971, p. 930.

NEMATOTHECIUM HANSFORDII SP. NOV.—A NEW GENERIC RECORD TO INDIA

DURING the course of mycological survey of Maharashtra State, a hyperparasitic fungus parasitizing sooty mold affecting *Memecylon edule* L. was collected. The sooty mold was identified as *Meliola memecyli* Syd. and the hyperparasite was found to belong to the Ascomycetous genus *Nematothecium*[1] Syd.—a well known hyperparasite on sooty molds. A review of literature[2] revealed that this genus was not previously reported from India. Furthermore, the present material on critical comparison with holotypic materials of *Nematothecium vinosum* Syd. and *N. asterinae* Hansf., which are the only two species of *Nematothecium* so far reported, was found to be significantly distinct in respect of dimensions of asci and ascospores, (*vide* Table I) besides being on a new host, *i.e. Meliola memecyli.*

The present fungus, therefore, is placed under a new taxon namely *Nematothecium hansfordii.* The specific epithet is named in the honour of Dr. C. G. Hansford, in recognition of his outstanding contributions to the field of mycology. The proposed new species is described below:

Nematothecium hansfordii sp. nov. (Fig. 1).

Fungus parasitic on colonies of *Meliola,* hyphae brownish pink, distinctly septate, 3–4 μ broad.

Perithecia numerous, aggregated within colonies of *Meliola,* made up of 2 layers of wall, outer dark brown, plectenchymatous; inner light brown, measuring 200–260 × 250–330 μ.

Asci in basal umbellate cluster, cylindrical, slightly curved, shortly stipitate; measuring 90–115 × 12–16 μ, paraphysate.

Paraphyses numerous hyaline to subhyaline simple and filiform.

Ascospores subfiliform to filiform, mostly sigmoid, curved at both the ends, 5–6 septate, olivaceous gray; measuring 82–105 × 4–6 μ.

TABLE I

Name of the Species	Host	Asci measurement	Ascospore measurement
1. *Nematothecium vinosum* Syd. (Type species)	*Meliola* sp.	65–90 × 10–14 µ	60–75 × 3–4 µ
2. *N. asterinae* Hansf.	*Asterolibertia burchelliae*	18–23 × 4–7 µ	16–20 × 1 µ
3. *N. hansfordii* sp. nov.	*Meliola memecyli*	90–115 × 12–16 µ	82–105 × 4–6 µ

On colonies of *Meliola-memecyli* Syd.
Holotype—AMH-2628.
Type loctlity—Khandala (M.S.).

Nematothecium hansfordii sp. nov. (Fig. 1).

Fungus in plagulis *Melioliae* parasitans ex hyphis, bruneo = roseum. Hyphae distincte septatae, 3–4 µ latum.

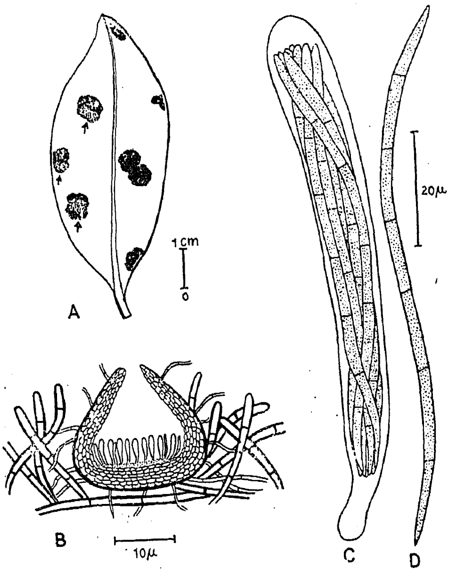

FIG. 1. *Nematothecium hansfordii* sp. nov. A, Habitat; B, V.S. of Perithecium; C, Ascus; D, Ascospore.

Perithecia numerosa, gregaria in plagulis *Melioliae;* paries bistratosi, exterium fusce-brunneus, plectenchymaticus, interius pallido brunneus; magnit 200–260 × 250–330 µ.

Asci in basalis umbellati fasciculi, cylindraceo-curvatiae, brevi-stipitatae; magnit 90–115 × 12–16 µ, paraphysaliae.

Paraphyses numerosae, hyalinae vel subhylinae, simplices et filiformes.

Ascosporae subfiliformae vel filiformae, pleurumque signoideacurvatiae, ad biden apicem, 5–6 septate, olivaceo-griscae; magnit 82–105 × 4–6 µ.

On plagulis *Meliola memecyli* Syd.
Holotype—AMH-2628.
Typus locus—Khandala (M.S.).

The authors are grateful to Prof. M. N. Kamat, Head, Department of Mycology and Plant Pathology, for valuable suggestions, to Dr. G. B. Deodikar, Director, for library and laboratory facilities and to Dr. Mrs. A. Pande, for Latin rendering. Thanks are also extended to the Curators of Herbaria at Kew and Stockholm, for kindly making the holotypic materials available.

Maharashtra Association for the Cultivation of Science,
Poona 411 004, *October* 13, 1975.
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J. G. VAIDYA.

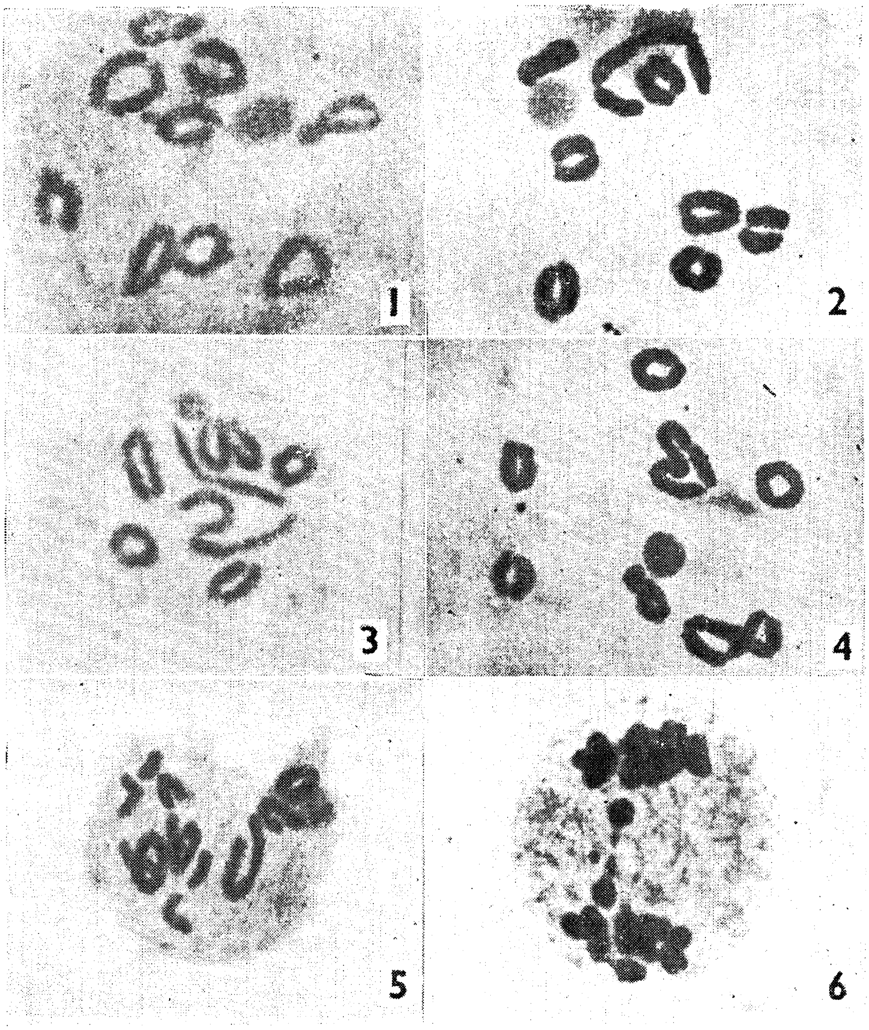
---

1. Hansford, C. G., *The Folicolous Ascomycetes, Their Parasites and Associated Fungi,* The I.M.I., Kew Surrey, Mycological Papers, 1946, No. 15, p. 240.
2. Mukherjii, K. G. and Juneja, R. C., *Fungi of India* (1962–72), Emkay, Pub., New Delhi, 1975, p. 245.

MAINTENANCE OF STRUCTURAL HETEROZYGOSITY IN THE F_2 GENERATION OF INTERSPECIFIC HYBRIDS OF ANNUAL CHRYSANTHEMUM

ANNUAL Chrysanthemums are of two distinct species. *Chrysanthemum coronarium* Linn., and *Chrysanthemum carinatum* Schousb. They are generally grown as winter garden annuals. Both the species are cross pollinated. Maintenance of interchange heterozygosity and its preponderance due to better reproductive fitness were noted in natural populations of these two species[1,2]. Nilsson[3] reported successful hybridization between these two species but a detailed analysis of this hybrid and segregants is not yet available. Interchange as well as inversion heterozygosity were observed in the hybrids. The objective of the present study was to analyse cytologically the F_2 plants of the hybrids between *C. coronarium* and *C. carinatum*.

Flower buds were fixed in Carnoy.s fixative for 24 hours and then preserved in 70% ethyl alcohol. Anthers of suitable size were squashed in propinocarmine using ferric chloride as mordant. Multivalent association in pollen mother cells were recorded from diakinesis and metaphase-1.



FIGS. 1–6. Photomicrographs of meiotic stages in F_2 plants of interspecific hybrids of Annual Chrysanthemum. Fig. 1. PMC with 9 II. Fig. 2. PMC with 7 II + 1 IV. Fig. 3. PMC with 6 II + 1 VI. Fig. 4. PMC with 5 II + 2 IV. Fig. 5. PMC with 5 I + 5 II + 1 III. Fig. 6. PMC with a dicentric bridge and an acentric fragment at anaphase-1.

Meiotic analysis of the F_2 segregants revealed the presence of structurally homozygous plants with nine bivalents as well as translocation heterozygotes (Figs. 1–5). Multivalent associations ranged from trivalent to decavalent configurations (Table I). This type of structural heterozygosity was also detected in the parents and hybrid[4]. Few univalents were found during meiosis in F_2 individuals. Univalents were also detected in the F_1 hybrid. At anaphase-1 a dicentric bridge and one acentric fragment were observed in the F_2 populations (6% cells) suggesting a paracentric inversion (Fig. 6).

The presence of structural heterozygosity in the F_2 generation indicates the adaptive role of these chromosomal rearrangements in annual chrysanthemum. Selection in favour of structural heterozygosity including that resulting from interchanges was reported earlier in several plant and animal species[5-8]. It is likely that chromosomal rearrangements found in the F_2 populations have a role in the establishment of adaptively favourable genes.

TABLE I

Distribution of chromosome associations in F_2 plants

Chromosome associations	Number of plants studied *C. coronarium* × *C. carinatum*	 *C. carinatum.* × *C. coronarium*
9 II	20	22
9 II + 1 IV	16	16
5 II + 2 IV	10	8
6 II + 1 VI	4	18
4 II + 1 IV + 1 VI	8	10
4 II + 2 III + 1 IV	2	..
1 II + 2 IV + 1 VIII	1	1
5 II + 1 VIII	6	2
3 II + 1 IV + 1 VIII	2	2
4 II + 1 X	..	1
5 I + 5 II + 1 III	2	1
4 I + 3 II + 2 IV	1	1
3 I + 6 II + 1 III	2	..
2 I + 6 II + 1 IV	1	1
Total No. of plants analysed	75	83

Such groups of genes would correspond to what Darlington and Mather[9] have called a 'super gene' or 'co-adapted gene complex' of Dobzhansky[10] and would be inherited as an unit. Univalents may occur either due to deysnapsis or failure of pairing between chromosomes of the two parental species. Desynapsis leading to univalents may be an isolating mechanism between the presently studied parental species as was reported in case of two species of *Oryza* by Shastry[11]. In paracentric inversions single crossover occurring within the inverted segments produce duplicate-deficient gamets resulting in sterility. Moreover, a paracentric inversion can bring co-adapted genes into closely linked situation without altering the karyotype symmetry. This preserves the internal balance of a species which might be prerequisite for its adaptation[12].

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---

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1. Rana, R. S. and Jain, H. K., *Heredity*, 1965 22, 21.
2. —, *Chromosoma*, 1965, 16, 477.
3. Nilsson, A., *Agri. Hort. Genet.*, 1964, 22, 160.
4. Chaudhury, B. K., *M.Sc. (Ag.) Thesis*, Faculty of Agriculture, University of Kalyani, West Bengal, (unpublished), 1969.
5. Darlington, C. D., *Proc. Roy. Soc.*, 1956, 145 B, 350.
6. Rees, H., *Evolution*, 1961, 15, 145.
7. John, B. and Lewis, K. R., *Heredity*, 1958, 12, 185.
8. Burnham, C. R., *Bot. Rev.*, 1956, 22, 419.
9. Darlington, C. D. and Mather, K., *The Elements of Genetics*, 1st Edn., Allen and Unwin, Ltd., London, 1952.
10. Dobzhansky, Th., *Genetics*, 1950, 35, 288.
11. Shastry, S. V. S., In : *Rice Genetics and Cytogenetics*, 1st Edn., Elsevier Publ. and Co., London, 1964.
12. Stebbins, G. L., *Adv. Genet.*, 1958. 9, 147.

KARYOTYPE OF *CYRTANTHUS MACKENII* HOOK. f.

THE genus *Cyrtanthus*, a member of Amaryllidaceae, comprises thirty species[10] of which many are endemic to South Africa. Although most of the species studied show $2n = 16$[1,1a,8,9], *C. sanguineus*, *C. ifafalily* and *C. obliquus* bear 18, 20 and 22[2,6] chromosomes respectively. Besides variation in chromosome number, the various species of *Cyrtanthus* differ markedly in their karyomorphology. In view of the above, the karyotype of *C. mackenii* obained from the Ooty Botanical Gardens was worked out.

In this species, the root stock is a tunicated bulb with several basal, linear leaves which are contemporary with the flowers. Scape is hollow, suberect and is subtended by 2–4 bracts bearing terminal orange red flowers. Stamens which are six in number with slender filaments and versatile anthers are inserted below the throat of the corolla tube. Ovary is inferior and trilocular with numerous campylotropous ovules on axile placentation. Style is filiform with three-lobed stigma. Seeds are many, black, flattened and winged.

Karyological analysis reveals the somatic chromosome number to be 16 of which two pairs are long, five are medium and one is short (Fig. 2). The chromosomes of the complement conform to the following types (Fig. 1).

1. A pair of long chromosomes ($12 \cdot 8\ \mu$) with median centromeres.
2. One pair of long chromosomes ($9 \cdot 2\ \mu$) with submedian primary constrictions.

3. A pair of medium chromosomes (7·6 μ) with submedian primary constrictions and secondary constrictions on the long arms.
4. One pair of medium sized chromosomes (7·2 μ) with submedian centromeres.
5. Two pairs of medium chromosomes (5·6 μ and 4·4 μ) with subterminal centromeres.
6. A pair of medium chromosomes (4·4 μ) with submedian centromeres.
7. One pair of short heteromorphic chromosomes (4·0 μ and 3·6 μ) with medium and subterminal centromeres.

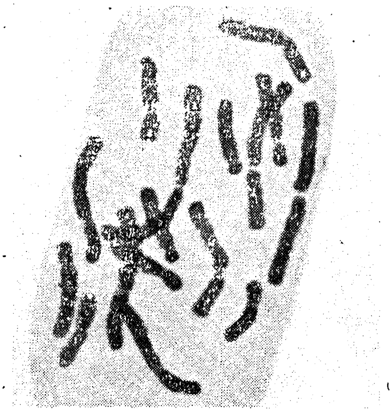

FIG. 1. Mitotic complement of *C. mackenii* showing the number $2n = 16$, × 1,100.

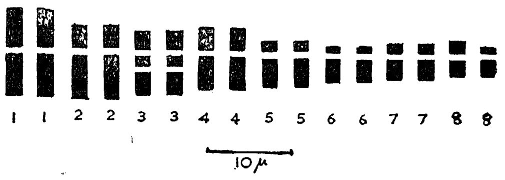

FIG. 2. Idiogram of *C. mackenii.*

Of the eight pairs of chromosomes, one pair possesses median centromeres, four submedian centromeres and of the remaining three pairs, two pairs have subterminal centromeres and the other pair shows heteromorphic chromosomes with median and subterminal centromeres.

The karyotype is symmetrical since the number of median and submedian chromosomes are more when compared to the subterminal ones[7].

From the present study, it is evident that the diploid chromosome number of *C. mackenii* is $2n = 16$ indicating 8 as the basic number. This observation is in conformity with those of Taylor[8] in *C. parviflorus* and Wilsenach[9] in eleven species of *Cyrtanthus,* except for the presence of one pair of heteromorphic chromosomes (8th pair) in the karyotype complement. It is likely that the origin of heteromorphic pair of chromosomes in the complement may possibly be due to the inversions or translocations which might have played a role in the evolution of the genus *Cyrtanthus* by bringing about changes in the morphology of the chromosomes.

From a reinvestigation carried out by Ising[1] and Wilsenach[9] on the three species, *C. sanguineus, C. ifafalily* and *C. obliquus,* they found the diploid number to the only 16 and regarded the reported number, $2n = 18$, 20 and 22 by Mookerjea[2] and Sato[6] to be erroneous. Since the phenomenon of polymorphism of chromosomes is common in the family Amaryllidaceae, as observed in the genera *Zephyranthes*[3], *Crinum*[4] and *Hymenocallis*[5], the reported occurrence of $2n = 18$, 20 and 22 numbers by Mookerjea[2] and Sato[6] need not be regarded as erroneous as they may represent different chromosomal races.

One of us (N. Lakshmi) is grateful to the C.S.I.R. (India) for financial assistance during the course of the work. Our thanks are also due to the Curator, Ooty Botanical Gardens.

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Guntur, A.P., *June* 13, 1975.

---

1. Ising, G., *Pl. Life,* 1962, **18**, 95.
1*a*. —, *Heriditas,* 1970, **65**, 1.
2. Mookerjea, A., *Caryologia,* 1955, **7**, 1.
3. Raina, S. N., and Khoshoo, T. N.. *Nucleus,* 1971 *b*, **14**, 23.
4. —, *Caryologia,* 1971 *c*, **24**, 217.
5. —, *La cellule,* 1971 *e*, **52** (3), 259.
6. Sato, D., *Cytologia,* 1938, **9**, 203.
7. Stebbins, G. L., *Chromosomal Evolution in Higher Plants,* Edward Arnold (Publishers) Ltd., London, 1971.
8. Taylor, W. R., *Amer. J. Bot.,* 1925, **12**, 104.
9. Wilsenach, R., *Cytologia,* 1963, **28**, 170.
10. Willis. J. C., *Dictionary of Flowering Plants,* Cambridge University Press, 1966.

A NEW RECORD OF *ARXIELLA TERRESTRIS* FOR INDIAN SOILS

WHILE studying the mycoflora of a clayey loam soil of the Botanical Gardens, Government Science College, Jabalpur, M.P., India, the authors encountered a hyphomycetous fungus which on identification[1], was found to be *Arxiella* Papendorf. The genus *Arxiella* with *A. terrestris* as the type species was erected by Papendorf[2] who isolated it from top soils and leaf litter of *Acacia Karroo* community in Transval, South Africa. Later, Ruscoe[3] described another species, *A. lunata* from the soil. The present isolate closely resembles *A. terrestris*. So far, the genus *Arxiella* has not been reported from India on any substrata. It is, therefore, the first record from the country. The morphological characters are briefly described below.

Colonies on potato dextrose agar growing slowly, compact, at first dull white, becoming olive green to greenish black with the age. Mycelium composed of much branched, septate, smooth, thick walled hyphae having cells often inflated, 2·5–3·0 μ in dia. Conidiophores 1- to many celled. Unicellular conidiophores of various shapes, 4·0–6·0 μ in dia. Multicellular conidiophores simple or branched, filamentous, all cells conidiogenous, sterigmata substending the conidium not left as a distinct tubular appendage on conidiophore. Conidia sessile or on short or slightly elongated sterigma produced singly or in short chains, reniform with the ends tapering obliquely and forming horn like projections with rounded apieces, with a distinct, widely truncate and slightly raised hilum near the base of one of the horns, thin walled, smooth, hyaline to faintly olivaceous, 6·4–16·5 × 3·0–4·6 μ (Fig. 1).

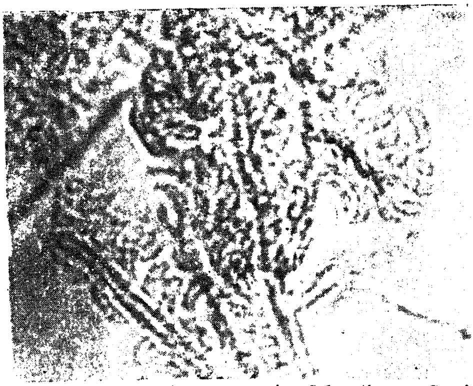

FIG. 1. *Arxiella terrestris*. Mycelium, Conidiophore and Conidia.

The culture has been deposited in the Science College Culture Collection, at Jabalpur.

The authors are thankful to Dr. M. B. Ellis of Commonwealth Mycological Institute, Kew, England, for confirming the identification to Dr. S. C. Agrawal, for help and to the Principal, Government Science College, Jabalpur, for the laboratory facilities.

Deparment of Botany, Govt. Science College, Jabalpur, M.P., *February* 6, 1974.
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P. D. AGRAWAL.

---

1. von Arx, J. A., *Genera of Fungi Sporulating in Pure Culture*, J. Cramer, Lehere, 1971, p. 288.
2. Papendorf, M. C., *Trans. Brit. mycol. Soc.*, 1967, 50, 69.
3. Ruscoe, Q. W., *Ibid.*, 1970, 55, 506.

VARIATION IN AMINO ACIDS IN THE GROWING CULTURE OF *ALTERNARIA ALTERNATA* (FR.) KEISSLER

A PROGRESSIVELY developing culture of *Alternaria alternata* (Fr.) Keissler obtained from leaf spot lesions of beet (*Beta vulgaris*) and grown in Czapeck's medium was chromatographically analysed for 30 days, at an interval of five days, for locating the presence of amino acids, both in the mycelial extract and culture filtrate. The methods adopted were the same as in an earlier investigation by Prasad and Chaudhary[11].

Comparative quantitative estimations are based on arbitrary gradations[12].

As would appear from Table I, mycelial extract (M.E.) of 5–10 days culture showed the presence of 7 amino acids. By the 15th day six more amino acids appeared. On the 25th day *l*-leucine/isoleucine and β-phenylalanine disappeared. Methionine appeared only once in the 15 day old mycelium. Lysine got lost after 15 days and after 25 days tyrosine, proline, glutamine and glutamic acids were also not found.

Very few amino acids were recorded in the culture filtrate (C.F.) of the pathogen. On the 5th day *dl*-alanine appeared. By the 10th day glutamine was observed and by the 15th, two more amino acids, (namely, homoserine and aspartic acids) were noticed. All the four, mentioned above, showed themselves only once in the culture filtrate after which they disappeared.

Serine, glycine, *dl*-alanine, tyrosine, aspartic acid, proline and lysine appearing in the mycelial extract, and *dl*-alanine in the culture filtrate, initiated the mycelial accumulation in the 5-day old culture.

With the increase in the amount of glycine, *dl*-alanine and aspartic acids in the mycelial extract, the fungal growth increased enormously by the 10th day. Significantly, the single appearance of a little amount of glutamine in the filtrate of this

TABLE I

Amino acids content of mycelial extract (M.E.) and culture filtrate (C.F.) of 5–30 days progressively growing culture of Alternaria alternata *(Fr.) Keissler*

Amino acid	Amount expressed in arbitrary units											
Days of incubation →	5		10		15		20		25		30	
	M.E.	C.F.	M.E.	C.F.	M.E.	C.F.	M.E.	C.F.	M.E.	C.F.	M.E.	C.F.
Serine ..	++	–	++	–	++	–	++	–	++	–	++	–
Glycine ..	+	–	++	–	+	–	+	–	+	–	+	–
dl-Alanine ..	++	+	+++	–	++	–	++	–	+	–	+	–
Tyrosine ..	++	–	+	–	+	–	+	–	+	–	–	–
Aspartic acid ..	+	–	++	–	++	+	++	–	+	–	+	–
Proline ..	+	–	+	–	++	–	++	–	+	–	–	–
Lysine monohydrochloride	+	–	+	–	+	–	–	–	–	–	–	–
Glutamine ..	–	–	–	+	+++	–	++	–	++	–	–	–
Homoserine ..	–	–	–	–	++	–	++	–	+++	–	–	–
Glutamic acid ..	–	–	–	–	++	–	++	–	++	–	–	–
Methionine ..	–	–	–	–	+	–	–	–	–	–	–	–
l-Leucine/isoleucine ..	–	–	–	–	++	–	++	–	–	–	–	–
β-phenylalanine ..	–	–	–	–	++	–	++	–	–	–	–	–
Dry weight of the fungal mat (in mg)	101		888		1035		1230		1395		1510	

age group of culture was, to a degree, also responsible for the sudden spurt of exuberant mycelial growth.

The appearance of glutamine, homoserine, glutamic acid, methionine, *l*-leucine/isoleucine and *β*-phenylalanine after 15 days of growth in the mycelial extract also raised the mycelial output. Thereafter, the mycelial growth increased uniformly in a linear form, with the days of incubation and irrespective of any alteration in the amount of the amino acid.

Qualitative and quantitative variations in the amino acid pool exist in the cultures of different fungi[3]. Mycelial extracts of *Pellicularia filamentosa, Pythium, Daldinia concentrica* and *Fusarium udum* showed the presence of twelve[10], fifteen[5], twelve[6] and fifteen[13], different amino acids respectively; in *Alternaria alternata,* as many as thirteen amino acids were detected.

The presence of aspartic and glutamic acids, the absence of *l*-cystine, and almost negligible record of methionine in the mycelium are similar to the observations made with 8 pathogenic species of *Phyllosticta*[2]. Alanine and lysine, found in *A. alternata,* have also been traced in 8 of *Fusarium* species[14]. A larger alanine and aspartic acid pool gets built up in 10–20 days old mycelium. In fact, this was true of *Penicillium roquefortii*[8] and *Fusarium moniliforme*[1], as well. As for its peculiar feature, the alanine and the aspartic acids, after showing an incerase in their amount between 5 and 10 days and also after remaining constant for 10 to 20 days, abruptly dropped down between 20 and 30 days. This is in marked contrast with the feature shown in *F. moniliforme*[1].

Further traits with regard to the mycelial extract of *A. alternata* are noted below: proline appeared and continued to be present, first in small amounts, and then increasing progressively for 5–20 days; later within 15–30 days, homoserine was detected, accumulating in the largest quantity by the 25th day; glutamine was formed within 15–25 days, first in large amounts and later in slightly small amounts; and finally, leucine/isoleucine and *β*-phenylalanine appeared within 15–20 days in good amounts. This, however, is not in accordance with the findings recorded earlier[1,2,14].

Venkata Ram[13] could register only 15 amino acids in *F. udum,* and Prasad and Chaudhary[11] detected all the 21 by the 15th day. Out of these 21 amino acids, 13 appeared in *A. alternata* also by the 15th day. Also, *β*-phenylalanine appeared simulately by the 15th day, as found in *F. oxysporum* f. *udum*[11].

Detection of amino acids in culture filtrate of different *Fusarium* species has been reported earlier[7,9,13-14]. According to Mahadevan and Caroselli[7] younger culture filtrates of *F. oxysporum* f. *lycopersici* would show a larger amino acid pool than the older ones. But, here, no such co-relation could be established, as the number of amino acids detected in the culture filtrate of *Alternaria alternata* was small and the quantity meagre. Our observations are similar to those of Bilai and Zakordonets[1] and Prasad and Chaudhary[11]. Alanine, homoserine and aspartic acids found here were among the five and seven amino acids appearing in the culture filtrate of *F. axysporum* f. *udum*[11], and *F. lycopersici*[4]. Glutamine was found in addition to the three mentioned above.

The authors extend their thanks to Prof. J. P. Sinha, Head of the Botany Department, Ranchi University, for the facilities provided to them.

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---

1. Bilai, V. I. and Zakordonets, L. A., *Mikrobiol. Zh. Akad. Nauk. U.S.S.R.*, 1965, 27, 3.
2. Bilgrami, K. S., *Flora* (*Alg. Bot. Ztng.*), 1963, 153, 49.
3. Chandra, S. and Tandon, R. N., *Ibid.*, 1962, 152, 531.
4. Flück, V. and Richele, K. H., *Phytopath. Z.*, 1955, 24, 455.
5. Hines, R. B., *Mycologia*, 1961, 52, 378.
6. Joshi, G. T. and Patwardhan, P. G., *Indian Phytopath.*, 1972, 25, 143.
7. Mahadevan, A. and Caroselli, N. E., *Phytopath. Z.*, 1966, 55, 181.
8. Meyers, E. and Knight, S. G., *Mycologia*, 1961, 53, 115.
9. Natarajan, S., *J. Indian bot. Soc.*, 1958, 37, 233.
10. Ogura, H., Akai, S. and Sato, T., *Ann. Phytopath. Soc. Japan*, 1961, 26, 31.
11. Prasad, M. and Chaudhary, S. K., *Phytopath. Z.*, 1973, 78, 147.
12. — and —, *Ibid.*, 1974, 81, 339.
13. Venkata Ram, C. S., *Proc. natl. Inst. Sci. India*, 1956, 22 B.
14. Zakordonets, L. A., *Mikrobiol. Zh. Akad. Nauk. U.S.S.R.*, 1965, 27, 6.

LAUNAEA MOSAIC—A NEW VIRUS DISEASE FROM INDIA

DURING the course of survey of virus diseases in 1973, a mosaic disease was observed on *Launaea asplenifolia* Hook.—A common weed in Aligarh. The characteristic features of diesease are severe mosaic mottling followed by reduction in leaf size. The plants remain stunted.

Under glass house conditions the virus was found to be readily transmissible by sap, only to the plants belonging to families Amaranthaceae, Chenopodiaceae, Compositae and Solanaceae. The details about symptoms and host reactions are given in Table 1 and Fig 1.

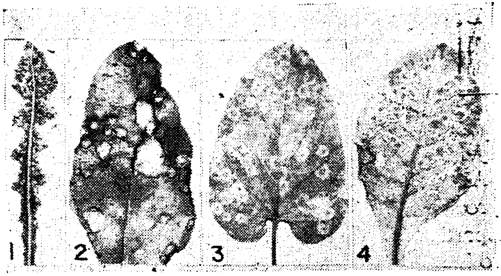

FIG. 1. Virus infected leaf of 1. *Launaea asplenifolia* Hook., 2. *Spinacia oleracea* L., 3. *Nicotiana glutinosa* L., 4. *N. tabacum* L. var. Samsun NN.

The virus did not infect 70 plant species belonging to 20 families nor could the virus be recovered on back inoculation tests.

Three species of aphids, *viz.*, *Aphis gossypii* Glov., *Brevicoryne brassicae* Sulz. and *Myzus persicae* L. in lots were given an initial starvation of 2 hours followed by ½ hour feeding on diseased plants and inoculation feeding of ½ hour on healthy plants. Only *A. gossypii* Glov. and *M. persicae* Sulz. transmitted the virus successfully while *B. brassicae* L. failed to transmit the virus.

The Launaea mosaic virus (LMV) in crude sap could tolerate heating at 60° for 10 minutes, but not at 65° C. The virus was found to be infective at a dilution of 10^{-3} but not 10^{-4}. The longevity *in vitro* of the virus in crude sap was found to be 36 hours at room temperature (20–25° C), but was 72 hours at 10° C.

A review of literature revealed that no virus has been reported on *Launaea asplenifolia* Hook. However it was used as a differential host for the leaf curl disease of tobacco by Pruthi and Samuel[1]. They reported that the leaf curl disease of tobacco was responsible to infect *Lycopersicon lycopersicum* (L.) Karsten, *Ageratum conyzoides* L., *Crotolaria juncea* L., *Euphorbia hirta* L., *Launaea asplenifolia* Hook., *Solanum nigrum* L., *Sida cordifolia* L., *Scoparia dulcis* L., *Veronica cineria* Boiss. and Bal. and *Zinnia elegans* Jacq. But the virus under the present study did not infect the above plants except *L. lycopersicum* (L.) Karsten. and *Zinnia elegans* Jacq. In spite of this the LMV produced local lesion on *Chenopodium album* L., *C. amaranticolor* Coste. and Reyn. and *C. murale* L., while leaf curl disease of tobacco failed to produce local lesion on the above three species. It is also quite different in its host reaction, symptomatology

TABLE I

Host range of Launaea mosaic virus (LMV) isolated from Launaea asplenifolia *Hook.*

Families/Latin name of the species inoculated	Symptoms		Back inoculation test
	Local	Systemic	
Amaranthaceae			
Achyrenthes aspera L.	Nll	Vc, N, Ld, S	+
Amaranthus caudatus L.	Cp	Vc, M, Ld, S	+
A. hypochondriacus L.	..	Vy, M, Ld, S	+
A. leucocarpus S. Wat.	Cp	N, Bm, Ld, S	+
Chenopodiaceae			
Beta vulgaris saccharifera L. var. Bushmona	Cs	Vy, N, M, S	+
Chenopodium album L.	Ll	..	+
C. amaranticolor Coste. and Reyn.	Ll	..	+
C. murale L.	Ll	..	+
Spinacia oleracea L.	Nll	..	−
Commelinaceae			
Commelina nudiflora L.	..	Ym, S	+
Compositae			
Zinnia elegens Jacq.	..	Vy, M, S	+
Solanaceae			
Datura stramonium L.	..	Vc, M, Ld, S	+
D. metel L.	..	Vc, M, Ld, S	+
D. alba Nees	..	Vc, M, N, Ld, S	+
Lycopersicon grandulosum Muller.	..	Vc, M, Ld, S	+
L. lycopersicum (L.) Karsten.	..	Vc, M, Ln, S	+
Nicotiana glutinosa L.	Dwnr	Vy, M, S	+
N. rustica L.	..	Vc, M, S	+
N. tabacum L. var. Harrison's Special	N	Vc, M, Ld, S	+
N. tabacum L. var. Samsun NN	..	Vc, M, Ld, S	+
N. tabacum L. var. White Burley	..	Vc, M, Ld, S	+
Petunia hybrida Vilm.	..	Vc, M, Ld, S	+
Solanum melongena L. var. Manjrigota	..	Vc, M, S S	+

+ and − indicate positive and negative results on back inoculation tests.

Abbreviations: Bm = Bare midrib; Cp = Chlorotic patches; Cs = Chlorotic spots; Dwnr = Double wall necrotic rings; Ld = Leaf deformation; Ln = Leaf necrosis; Ll = Local lesion; M = Mosaic; N = Necrosis; Nll = Necrotic local lesions; S = Stunting; Vc = Vein clearing; Vy = Vein yellowing; Ym = Yellow mosaic.

and physical properties with that of leaf curl disease of tobacco.

It appears, therefore, that the virus under study is a new virus disease of *Launaea asplenifolia* Hook. The name Launaea mosaic virus has tentatively been proposed for the virus causing mosaic disease of *L. asplenifolia* Hook.

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1. Pruthi, H. S. and Samuel, C. K., *Indian J. agric. Sci.*, 1942, **12**, 35.

SHORT SCIENTIFIC NOTES

Colorimetric Estimation of 2, 4, 5-Triamino-6-Hydroxypyrimidine

2, 4, 5-Triamino-6-hydroxypyrimidine (TAP) is an important intermediate in the synthesis of folic acid. Its purity is usually determined by iodometry[1]. We have found that it reacts with ninhydrin to give a purple colour showing an absorption maximum at 555 nm. The colour is stable for 40 minutes and obeys Beer's law in the concentration range of 2·4 to 22 μg/ml of TAP sulphate (TAPS). Optimum conditions for colour development and estimation of TAPS have been worked out and are reported here. The procedure is simple, fast and accurate. Procedure—About 60 mg of TAPS was weighed accurately into a 250 ml calibrated flask and dissolved in 200 ml of warm water. The solution was allowed to cool to room temperature, 20 ml of 10% (v/v) hydrochloric acid was added and the final volume was made up with water. One ml of the solution was placed in a 25 ml volumetric flask and 10 ml of isopropanol and 2 ml of ninhydrin reagent (3% w/v solution in methyl cellosolve) was added. The reactants were mixed well and the resulting coloured solution diluted to mark with isopropanol. Absorbance of the colour was measured at 555 nm against a reagent blank on a Bausch and Lomb Spectronic-20 Spectrophotometer. A standard solution of TAPS, prepared from a sample assaying 100·0% by iodometry exactly in the same manner as described above, was run simultaneously. The absorbance of the standard colour was used to calculate the purity of TAPS sample. It has been observed that solutions of TAPS on storage show slight change in colour and hence it is advisable to prepare standard as and when it is required.

Five batches of TAPS from our plant have been analysed by the proposed method and the results are compared with those obtained by iodometry (Table I).

TABLE I

Analysis of TAPS by iodometry and colorimetry

Sl. No.	% content of TAPS Colorimetry	Iodometry	Deviation
1.	100·5	99·7	+0·8
2.	100·0	100·0	..
3.	100·8	100·3	+0·5
4.	100·3	99·7	+0·6
5.	99·9	99·3	+0·6

The method is being extended to the estimation of folic acid which gives TAP on reductive cleavage by heating with zinc and hydrochloric acid.

Quality Control Laboratories,
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1. Devianthnin, V. A., *Nature of Chemical Analysis in the Process of Vitamin Manufacture*, Ministry of Medicine, Mascow, 1964, p. 193.

Foot and Mouth Vaccination Stress in Red Dane, Jersey and Murrah Bulls

Increased use of exotic bulls to improve our livestock has lead us to employ recent technological advances for protecting them and their progeny against widely prevalent infectious diseases. Preventive vaccinations against such diseases have been reported to increase spermatozoal abnormalities in breeding bulls (Venkataswami and Rao[1], Narasimhan *et al.*[2] and Rao[3]) resulting in testicular damage (Rao and Venkataswami[4]). The results of the present investigation would reveal the effect of foot and mouth disease vaccination on details of sperm abnormalities and preservability of semen in exotic (Red Dane and Jersey) and indigeneous (Murrah) bulls.

2 Red Dane, 3 Jersey and 4 Murrah bulls were vaccinated, each with 40 ml polyvalent foot and mouth vaccine (I.V.R.I., Izatnagar). Their semen ejaculates, each at a week's interval, for 15–21 and 61–75 pre-and post-vaccination days respectively, were evaluated for sperm abnormalities (head, middle piece, tail and total). These semen samples of bull (Red Dane and Jersey) and buffalo bull (Murrah) were also extended in egg yolk citrate and egg yolk glucose bicarbonate dilutors (Saxena[5]) respectively and stored at 3°–5° C. These were evaluated at every 24 hourly interval for 168 hrs for spermatozoal motility (progressive and per cent) *vis-a-vis* its suitability for artifical insemination (A.I.).

The percentage of abnormal spermatozoa increased significantly during 16–30, 0–45 and 0–45 days, post vaccination in Red Dane, Jersey and Murrah bulls respectively. Majority of the abnormalities were found in the mid piece region (mainly cytoplasmic droplets and coiled/kinked) in all the three species of animals. It indicated that maturation process was somehow adversely affected. The preservability of spermatozoa (number of days semen found suitable for A.I.) was also adversely affected during the post-vaccination period. It indicated close relationship between incidence of sperm abnormalities and semen preservation. Details will be reported later.

We are thankful to Dean, College of Veterinary Medicine and Director, Experiment Station, G. B. Pant University of Agriculture and Technology, Pantganar, for prôviding the necessary facilities.

G. B. Pant University of Agriculture and Technology,
Pantnagar,
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---

1. Venkataswami, V. and Rao, V. J., *Indian Vet. J.*, 1970, 47, 23.
2. Narasimhan, K. S., Krishnan, A. R. and Quayam, S. A., *Ibid.*, 1970, 47, 19.
3. Rao, A. V., *Ibid.*, 1974, 51, 74.
4. Rao, V. J. and Venkataswami, V., *Ibid.*, 1971, 49, 1101.
5. Saxena, V. B., *M.V.Sc. Thesis*, Agra University, Agra, 1965, p. 23.

Hydrocyanic Acid Concentration at Different Ages of High Yielding Varieties of Sorghum

Sorghum grain forms a staple human food while the nutritious green fodder is of great value to animals. The main objection for the extensive use of this fodder is the presence of cyanogenic glucoside which on hydrolysis yields HCN, a toxic principle to the livestock at higher concentrations. The HCN content of sorghum plant as affected by various factors like variety, nitrogenous fertilizer application, age and stage of the crop and irrigation has been well studied[4,5]. The present study was undertaken to find out the amount of HCN during different ages of crop growth.

A field trial, under irrigated conditions, was conducted with three high yielding varieties of sorghum, viz., CSH. 5, CS. 3541 and 699 adopting a uniform fertilizer schedule of 100 N, 80 P_2O_5 and 60 K_2O kg/ha. The samples were collected at 30, 45, 60, 75 and 80th days for analysis. In all the varieties, flowering was observed in 60 days. The HCN content was estimated by colorimetric method[6] using the standard curve as described by Gilchrist *et al.*[3].

HCN content during various age of crop is presented in Table I.

The HCN concentration ranged from 485 to 10 ppm. A gradual reduction in the HCN concentration was seen in all the three varieties tried and a substantial reduction was noted after 75 days. Similar observations were also reported elsewhere[5,7].

Table I

HCN concentration during different ages of crop growth

Age in days	HCN (ppm) on fresh weight basis		
	CSH·5	CS·3541	699
30	465	485	355
45	210	440	245
60	125	135	125
75	30	50	60
80	30	10	30

The HCN concentration after 75 days is well below the toxic limits as suggested by Bolhuis[1] and de Bruijn[2]. They classify those which contain less than 50 mg HCN/kg fresh peeled tuber of cassava as innocuous.

From the present study it can be concluded that the fodder can safely be utilised, when cut after 75 days.

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---

1. Bolhuis, G. G. Neth, *J. Agric. Sci.*, 1954, 2, 176.
2. de Bruijn, G. H., *Etude du Caractere Cyanogenetique du Manioc.*, Vermam and Zonen, Wageningen, 1971.
3. Gilchrist, D. G., Lueschen, W. E. and Hittle, C. N., *Crop. Sci.*, 1967, 7, 267.
4. Hogg, P. G. and Ahlgreen, H. L., *J. Am. Soc. Agron.*, 1942, 34, 199.
5. Hukkeri, S. B., Rajput, R. K., Mukherjee, R., Singh, V. and Shukla, N. P., *Indian J. Agrl. Sci.*, 1972, 42, 648.
6. Indira, P. and Sinha, S. K., *Indian J. Agric. Sci.*, 1969, 39, 1021.
7. Shaikh, G. A. and Zende, G. K., *Ibid.*, 1971, 41, 456.

New Records of a Rare Bonellid *Ikedella misakiensis* (Ikeda)

A bonellid worm, *Ikedella misakiensis* (Ikeda) belongs to phylum Echiuroidea and is of much academic interest as it exhibits sexual dimorphism. The only available information regarding the occurrence and distribution of this species is from

Pirotan Island (Lat. 22° 36′ 12″ N Long. 69° 56′ 58″ E) in India (Gideon *et al.*[1]).

During the course of coral survey in the neighbourhood of the Gulf of Kutch, the author came across several specimens of this species from different localities for the first time (Table I). The specimens are deposited in the Museum of Marine Biological Research Station, Port Okha. The present records extend the distributional range of the species to Indian waters.

TABLE I

New records of Ikedella misakiensis (*Ikeda*)

Sl. No.	Locality	Specimen observed
1.	Mithapur in Arabian Sea	9
2.	North-eastern side of Paga reef	1
3.	South-western side of Pashu Island	7
4.	North-western side of Baida Island on Bural reef	8
5.	South-western side of Nora Island on Bural reef	1

The author is thankful to Shri Moosa Raza, Fisheries Commissioner, for the facilities and for permitting to publish this note. Special thanks are also due to Shri P. P. Bhanderi and Shri A. B. Chauhan, for field assistance.

Marine Biological
Research Station,
Port Okha, *December* 29, 1975.

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---

1. Gideon, P. W., Menon, P. K. B., Rao, S. R. V. and Jose, K. V., *J. Bombay nat. Hist. Soc.*, 1957, 54, 690.

Wet Rot of Brinjals (*Solanum melongena* L.) Caused by *Choanephora cucurbitarum* (Berk & Rev.) Thaxter

During September 1974, a serious fruit rot of *Solanum melongena* L. (Brinjals) was observed in Kankarkhera village of Meerut District. On examination it was found to be affected by *Choanephora cucurbitarum* (Berk and Rev.) Thaxter. This fungus has been reported on the species of *Capsicum, Colocasia, Cassia, Gossipium, Hibiscus, Cajanus, Vigna* (Dastur[2], Sinha[5], Chowdhury and Mathur[1], Mitter and Tandon[4], Misra and Mehra[3] and Wilson and Jose[6]), respectively. It has been reported on the decomposing leaves of *Solanum melongena* L. but as far as the authors are aware, wet rot of fruits has not been reported.

The disease appears as water soaked spots on the leaves, flowers and fruits. Ultimately the fruits become soft and pulpy and are found to be covered with white shining cottony growth of the fungus. Careful examination of the specimen reveals the presence of black pin head sized structures. The pathogen was transferred to potato dextrose agar medium (P.D.A.) and yielded both conidial and sporangial stages.

The authors are grateful to Dr. M. R. Sharma, for encouragement and to Dr. V. Singh, for providing the laboratory facilities.

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Meerut, *September* 4, 1975.

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---

1. Chowdhury, H. P. and Mathur, R. S., *Sci. and Cult.*, 1935, 3, 423.
2. Dastur, J. F., *Ann. Bot.*, 1920, 34, 399.
3. Misra, D. P. and Mehra, R. C. S., *Indian Phytopath.*, 1969, 22, 515.
4. Mitter, J. N. and Tandon, R. N., *Proc. Indian Acad. Sci.*, 1937, 6, 194.
5. Sinha, S., *Ibid.*, 1940, 11, 162, 167.
6. Wilson, K. I. and Jose, P. C., *Indian Phytopath.*, 1968, 21, 130.

Record of a New Alternate Host Plant of Rice Leaf Miner, *Cerodontha oryzvora* Spencer.

Cerodontha oryzivora Spencer was observed from 1971 onwards causing damage in early stage of rice crop in Gujarat. The incidence of this pest is observed in nursery and after transplanting upto boot stage as the maggot feeds only on the tender leaves.

It was also noticed that the pest was present on the wheat (*Triticum durum* L.) which followed paddy cultivation during the month of October-November, 1974 indicating the migration of the insect. This area having monocropping pattern of raising only paddy crop in large acreage followed by *rabi* wheat as rotation year after year, may have brought about the host cross over of the pest.

The data collected showed of incidence of leaf damage (10–15%) on the wheat field. The spraying of Methyl demeton 0·03% and Dimethoate 0·08% gave good results.

The authors are thankful to the Director, Commonwealth Institute of Entomology, London, for the identification of the pest.

Main Rice Research Station,
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Nawagam,
September 1, 1975.

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REVIEWS AND NOTICES OF BOOKS

Hand-Book of Metal Ligand Heats and Related Thermodynamic Quantities. (Second Edition). By J. J. Christensen, D. J. Eatough and R. M. Izatt. (Marcel Dekker, Inc., N.Y.), 1975. Pp. vii + 495. Price $ 25.00.

This book is the enlarged version of the first edition. There are 1083 separate ligand entries as against 514 in the first edition which appeared in 1970. The literature covered is upto early 1974 and the number of entries suggest a real growth.

In the first seven pages, the authors have provided information relating to the convenient and proper use of the tables. The tables list the ligands, metal ions, thermodynamic quantities such as log K, Δ H, Δ S and Δ Cp and appropriate reaction conditions and pertinent references. The metal ions are arranged groupwise and the ligands alphabetically. The book carries four indexes, empirical formula index, elements in alphabetical order, synonyms for ligand and references. The references are arranged yearwise and the authors alphabetically. This system of indexing provides a convenient source of cross referencing. However, in none of the indexes, page numbers have assumed any significance.

This book is intended as a companion volume to the already published tables of stability constants of A. E. Martell and L. G. Sillen, and related books. This book will serve as a reference source for workers in the area of thermochemistry and thermodynamics of metal-ligand interaction and valuable for researchers in the field of coordination chemistry. The authors have done a creditable job of summarizing and compiling good number of thermodynamic quantities. This will be a useful addition to the Library. V. KRISHNAN.

Composition of Cosmic Radiation. By Krishna M. V. Apparao. (Gordon and Breach Science Publishers, London, W.C. 2), 1975. Pp. x + 86. Price £ 5·40.

Composition of Cosmic Radiation by K. M. V. Apparao, is more appropriate as a review article than as a book. Even as a review article, it essentially focusses the contributions from the Tata Institute of Fundamental Research and does not objectively treat the subject-matter. Whereas some amount of bias is inevitable, it is regrettable to see this attitude being taken to the extreme even in the case of a book.

In trying to make his monograph a comprehensive one, the author has touched upon various subjects such as solar wind, interplanetary magnetic field, solar cosmic rays, theories of modulation and cosmic X and Gamma rays. It is a pity that, in doing so, he has resorted to a pedestrian treatment which can lead to erroneous impression of the present status of the subject. Really significant contributions in these areas by Wolfe *et al.*, Hundhausen *et al.*, Gringauz *et al.*, McCracken and Rao, to name only a few, have been completely left out. The only two chapters which are presented in somewhat an acceptable form are Chapters II and IV which deal with the observational material on the composition of cosmic rays near the earth and the cosmic radiation in interstellar space respectively.

The recent evidence obtained from Pioneers 10 and 11 spacecrafts have considerably changed the picture of low energy cosmic rays near the earth. Whereas non-inclusion of this aspect is not taken against the author, since the book was presumably written before the new evidence was available, the value of the book itself becomes questionable when considered together with the way the subject-matter has been treated in various chapters. U. R. RAO.

Polymer Chemistry—An Introduction. By Malcolm P. Stevens. (Addison-Wesley Publishing Company, Inc., Reading, Massachusetts), 1975. Pp. xv + 458. Price $ 25.00.

The book *Polymer Chemistry—An Introduction* consists of 3 parts and an appendix. Part I deals with the Introduction to Polymers in 4 chapters. Chapter 1 deals with the basic principles starting with an introduction and historical development, definitions, and the basic principles of polymerisation and kinetics. Chapter 2 deals with the different molecular weights and briefly mentions about the methods of their measurement. "Chemical Structure and Polymer Properties" is the subject matter of Chapter 3 while "Evaluation, Characterisation and Analysis of Polymers" is the subject of Chapter 4.

'Vinyl Polymers' is the topic of Part II which again consists of 4 Chapters. Free radical and ionic polymerisation, polymerisation with complex coordination catalysts and reactions of Vinyl Polymers are the topics lucidly covered in these 4 chapters.

Part III covers non-vinyl polymers in Seven Chapters. polyethers, polysulphides and related polymers, polyesters, polyamides and related polymers, phenol, urea-, and melamine-formaldehyde resins, inorganic-organic polymers, miscellaneous organic polymers, and natural polymers are the topics briefly covered in these seven chapters.

As the author has said in the preface a course in *Polymer Chemistry* is being taught in many universities now at the graduate and undergraduate levels and there is a need for an introductory text. This book aptly serves this requirement. Since it lucidly covers all the topics at a fundamental level, it readily serves as a reference source for a practicing research chemist or for an industrial chemist with no prior training in polymer chemistry.

No book on polymers could be complete without a laboratory course or demonstration. Since a book of this type cannot include all the laboratory experiments, the author's forethought in including an appendix with sources of the laboratory experiments in polymer chemistry is commendable. The list of all the laboratory manuals, books and journal articles, covering various aspects of polymer synthesis and characterisation is included in this apendix which serves as a ready reference for the instructors and induces them to include demonstrations during the course of teaching.

V. KALPAGAM.

Theory of Non-neutral Plasmas. By Ronald, C. Davidson. (Addison-Wesley, Advanced Book Programme, Reading, Mass. 01867), 1974. Pp. xiii + 199. Price $ 15.00.

Till date books on the physical principles and mathematical models of plasmas have, without exception, assumed overall charge neutrality of these media. It is now well understood that overall charge neutrality is not a physical requirement for the existence of collective oscillations and shielding effects, the basic properties of plasmas, in many-body charged particle systems. Non-neutral plasma systems though amenable to theoretical treatment parallel to that of neutral plasmas, show many dramatic variations in behaviour under the similar electromagnetic fields. Non-neutral plasmas can therefore be treated as a subfield of the highly developed field of plasma physics.

The understanding of equilibria and stability of magnetically confined non-neutral plasmas is important in the study of plasma systems with large self fields, which play an important role in collective effect ring accelerators, generation and transport of intense high-current relativistic electron beams, and stripping and confinement of heavy ions in non-neutral electron clouds in both toroidal and mirror magnetic field configurations. Hence, this book gives an introduction of the theory of non-neutral plasmas and is timely in the developing subfield of plasma physics.

Professor Davidson, a well known plasma physicist and author (*Methods in Non-linear Plasma Theory*) has many important contributions in the theory of non-neutral plasmas to his credit. The present book certainly gives a lucid and well organized account of non-neutral plasmas.

The discussion of the motion of charged particles in the presence of a uniform magnetic field is followed by mathematical descriptions of non-neutral plasmas. Employing both the macroscopic and kinetic descriptions, equilibria and their stability are discussed for magnetically confined electron rich plasma columns, relativistic electron beams and magnetically confined, partially neutralized, relativistic electron rings. The author has painstatingly pointed out for each case the differences and similarities of the corresponding cases in neutral plasmas. This effort has made this book interesting both for the theoreticians and the engineers.

C. UBEROI.

A First Course in Applied Electronics (An Introduction to Microelectronic Systems). By W. Gosling. (Macmillan Press, Ltd., London). First Published 1975. Pp. viii + 162. Price £ 2.95 net.

In these days, linear integrated circuits are rapidly displacing discrete component electronics, in a variety of electronic equipment such as radio receivers and communication systems, instrumentation, automatic control, etc. The first step in miniaturization was the "Cardwood" assembly approach in which about hundred components were packed in a volume of one cubic inch which indicates the degree of miniaturization achieved employing conventional discrete components. In the semiconductor integrated approach, the design and construction of quite a large percentage of the components is integrated. Since the circuits are produced on a mass scale, the cost also is quite low with the result that practically all low power electronic circuits could come under integrated circuitry. The semiconductor integrated circuit is a basic amplifier unit. By the use of one or two conventional components along with the integrated circuit, it is possible to have circuits performing different operations. The integrated circuit contains systems of interconnected transitsors, diodes, resistors, capacitors just as discrete component circuits with the difference that the integrated circuit is formed completely within a single crystal of semiconductor silicon. The components are connected in a single series of processing operations. The electronics engineer has to accept the available mass produced integrated circuit and modify the circuit for his individual requirements by connecting additional discrete components.

It is against this background, that one has to view the utility of the present book. The book is mainly concerned with the application of integrated circuits to electronic systems. After considering the various types of amplifiers in Chapter 2, the author has pointed out variable dissipation amplifiers; and switching amplifiers find use in integrated circuits. In Chapter 3, there is the discussion of the equivalent circuit and frequency response of amplifiers. The determination of useful working region is illustrated with respect to bipolar transistors. The noise and the drift which set the lower limit for signal amplitude is discussed in Chapter 4. There is a very good discussion of both current and voltage feedback theory in Chapter 5. In Chapter 6, practical examples on the application of feedback, extending the bandwidth by the application of negative feedback, positive feedback in oscillatory circuits and bistable trigger circuits, reduction in distortion and stabilization of the working point. The last aspect is illustrated taking the example of microelectronic amplifier type 741 and single transistor amplifier. The case of unwanted positive feedback causing increase in gain, or even oscillation near the extremes of the pass-band, is discussed in Chapter 7. Since at present, linear integrated circuit amplifiers use mostly bipolar transistors, discussion of the integrated amplifier circuit is restricted to bipolar transistors. The 741–sophisticated operation amplifier is discussed in great detail. In addition the 733 type of amplifier which extends the frequency response to 100 MHz as against 1 MHz in 741 type is also discussed. The applications of 710–integrated amplifier is also discussed.

The book contains a very good introduction into integrated circuits. It enables the reader to design simple circuits. The book is a very welcome addition to the library of all students interested in integrated circuits. It is also useful to physicists and chemists who would like to have a working knowledge in this branch of Applied Electronics. 'Answers to questions' is a particularly welcome feature in this book.

R. S. Subrahmanya.

Cytogenetics and Plant Breeding (Sekharan and Sarathy). Revised and Enlarged by N. Krishnaswamy and N. Hrishi. Third Edition. (P. Varadachary and Co., Madras 600 001). 1975. Pp. 694. Price Rs. 37·50.

The issuance of a third edition of the popular text book on cytogenetics and plant breeding is to be welcomed. There is a wide need for a text book on these subjects which can be used by the students to follow the lecture courses in the Agricultural Colleges in India. It is evident that this book is being used as a text.

This book is more useful than similar texts from Western countries since it deals comprehensively with the crops grown in India and examples from Indian crop research are freely used. The text is large and comprehensive and would be of use even after graduation and hence worth owning. In comparison with one other Indian text on these subjects, this book is better in scope as well as accuracy. The eleven appendices would all be useful and the one giving the glossary is commended. Because of joint authorship there is unequal treatment of the topics. Currently it is fashionable to emphasis "Molecular Biology" and to ignore crops and crop production. This bias is not there, but the chapter on genes and structure of gene is superficial since the importance of biochemistry is not brought out. In comparison, the chapters on Sex Determination and Cytoplasmic Inheritance are elaborate.

The students may be required to learn fully only selected portions of the text; however, a comprehensive study would be of help to all those concerned with Indian Agriculture.

It would be possible to point out some examples of errors and omissions, but these are easily remediable. Though some illustrations are not clear, the students are bound to see living plants and specimens. Considerable care has been taken with style and expression, and this is to be commended under present circumstances in India. S. Sampath.

Award of Research Degree

Sri Venkateswara University, Tirupati, has awarded the Ph.D. degree in Physics to Shri E. Rajagopal; Ph.D. degree in Chemistry to Shri K. C. Mohan Rao, R. Venkataraman, Shri D. Bhaskar Reddy, Shri S. Jayarama Reddy, Shri G. Lakshminarayana; Shri M. V. Prabhakara Rao, Shri C. S. Dorai and Shri M. Radhakrishnaiah; Ph.D. degree in Botany to Shri P. Subrahmanyam and Shri A. S. Raghavendra. Ph.D. degree in Zoology to Shri K. Venkataramana Rao and Shri Y. Srinivasa Reddy; Ph.D. degree in Geology to Shri V. Damodar Reddy and Shri E. Doraswamy Naidu.

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PREPARATION AND ESTIMATION OF THIOSEMICARBAZIDE COMPLEXES OF PLATINUM AND PALLADIUM WITH CHLORAMINE-T AND DICHLORAMINE-T

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ABSTRACT

'Ionic' and 'neutral' complexes of thiosemicarbazide (TSC) with Pt and Pd, of the type $M(TSC)_2X_2$, and $M(TSC-H)_2$ respectively, (where X = Cl, Br, I, CN, CNS and NO_3) have been prepared from $H_2PtCl_6.xH_2O$ and $PdCl_2$. The ligand in the complexes undergoes oxidation with chloramine-T and dichloramine-T, with a 24-electron change per molecule of the complex. Volumetric methods for the estimation of the complexes have been proposed.

THIOSEMICARBAZIDE (TSC) is well known as a metal complexing agent and also finds application in the characterization of aldehydes, ketones and polysaccharides. Since a number of metal complexes of the compound are known, suitable analytical techniques are essential for estimating the compound in the free state as well as in the complexes. The methods for its estimation reported so far, based on its oxidation by alkali metal hypohalites[1,2] and lead tetra-acetate[3], are limited to the estimation of small quantities and are slow. Chloramine-T (CAT) and recently dichloramine-T (DCT) have been successfully used in assaying TSC and some of its metal complexes[4-6]. In the present investigations, we have reported the preparation of some 'ionic' complexes of Pt (II) and Pd (II) and their volumetric estimation by CAT and DCT. The communication also reports the oxidation of two 'neutral' complexes of Pt and Pd by these oxidants.

EXPERIMENTAL

E'Merck thiosemicarbazide was purified by recrystallization from aqueous solution. $PdCl_2$ and H_2PtCl_6. xH_2O (Johnson-Matthey Ltd., London) were used for preparing the complexes. Chloramine-T (Merck) was purified by the method of Morris *et al.*[7] An approximately decinormal solution was prepared and standardized by iodometry. DCT was prepared by the method of Jacob and Nair[8] and an approximately decinormal solution of the compound in glacial acetic acid was standardized by the iodometric method. UV spectra were obtained from Beckman DB spectrophotometer. IR spectra (KBr disc) were recorded on a Carl Zeiss UR-10 Infra red spectrophotometer.

Preparation of Complexes

Pt $(TSC)_2$ Cl_2.—This was prepared by adding dropwise an ethanol solution of $H_2PtCl_6 \cdot xH_2O$ to a hot saturated solution of the ligand in ethanol, in slightly beyond 1 · 2 molar ratio. The buff colored complex separated was then filtered, washed with hot ethanol and dried over P_2O_5 in vacuo. $Pt(TSC)_2Cl_2$ is readily soluble in water but is insoluble in alcohol.

$Pt(TSC)_2X_2$, where X = Br, I, CN, CNS and NO_3 was prepared by adding saturated solutions of the respective alkali halide, cyanide, thiocyanate or nitrate to the ice cold aqueous solutions of $Pt(TSC)_2.Cl_2$. The precipitate obtained after stirring was filtered and purified by recrystallization from aqueous solution.

Pd $(TSC)_2$ Cl_2.—A solution of TSC in 2 M HCl was added to a 2 M HCl solution of $PdCl_2$ in 2:1 molar ratio, with constant stirring. The yellow precipitate of the complex was filtered, washed with 0·1 M HCl and dried over P_2O_5 in vacuo.

The preparation of $Pd(TSC)_2X_2$ where X = Br, I, CNS and NO_3 was similar to the corresponding platinum complex described above.

The 'neutral' complexes $Pt(TSC-H)_2$* and Pd $(TSC-H)_2$* were precipitated as a silky blue solid and a greenish yellow solid respectively by the addition of 1 M ammonia to aqueous solutions of the corresponding chloride complex, until the solution becomes weakly alkaline.

The composition of the complexes was checked by elemental analyses. All the complexes were found to be diamagnetic, as expected for the square planar complexes. Conductivity measurements on aqueous solutions (10^{-3} M) of ionic complexes showed that they are 1:2 electrolytes.[9] UV spectra of the complexes show an intra ligand band around 43,000 cm^{-1}, while a charge transfer band (from sulphur to metal) is found around 37,000 cm^{-1}. A common band at 28,510 cm^{-1} is attributed to a d-d transfer, characteristic of square planar complexes[10].

Infrared spectra of the complexes showed that the N-H stretching frequencies around 3000 cm^{-1} in TSC are affected by complex formation[11]. Further, the strong band at 800 cm^{-1} in TSC attributed to pure C = S stretch[12] shifts to lower regions by as much as 100 cm^{-1} in the complexes. It can safely be assumed that the co-ordination of the metal ion takes place through both the nitrogen atom of the hydrazinic residue and sulphur atom of the ligand, as found in the case of zinc group complexes of TSC[11].

---

* (TSC–H) represents ($H_2NNCSNH_2$).

Some preliminary analytical investigations were carried out with the complexes. It was found that the ligand in the complexes was oxidized by CAT (with a 50–60% excess) at all pH. Oxidation was slow at $pH < 1$, faster in the pH range 1–3 and slower again at $pH > 5$. Oxidation was fastest at pH 4. Higuchi *et al.*[13] have shown that monochloramine-T (RNHCl, where $R = P\text{–}CH_3 - C_6H_4SO_2$), formed in acidified solutions of CAT, disproportionates to DCT and *p*-toluene sulphonamide and the disproportionation rate is maximum around pH4. The behaviour noticed with the oxidation of TSC complexes with CAT could probably reflect the high rate of disproportionation of the free acid RNHCl, which is also a fairly reactive species. Stoichiometric oxidation of the complexes took place within 30 minutes.

In oxidations with DCT, it was found that with a 50–60% excess of oxidant, the oxidation of the complexes (aqueous solutions or solutions in pH 4 buffer) was completed in 30 minutes, if an overall water content of about 10–15% was maintained in the reaction mixture. The rate of oxidation was found to be very slow with solutions of the complex in glacial acetic acid, the required stoichiometry being obtained after 16 hours[6].

The ligand TSC in the complexes is oxidized by CAT or DCT with a 24 electron change per molecule of the complex. It was noticed that the anions CN^- and CNS^- present in the complexes are also oxidized by the oxidants under these conditions, with a 4-electron change[14] and 16 electron change[14,15] per molecule of complex respectively. Allowance was made for this fact in calculating the amount of complex recovered.

RESULTS AND DISCUSSION

The stoichiometry of oxidation of the complexes can be represented as follows:

$$M(NH_2NHCSH_2)_2X_2 + 12\,RNClNa\ (\text{or } 6\,RNCl_2) + 12\,H_2O \rightarrow MX_2 + 12\,RNH_2\ (\text{or } 6\,RNH_2) + 2\,CO_2 + 2\,N_2 + 12\,NaCl\ (\text{or } 12\ HCl) + H_2SO_4 + (NH_4)_2SO_4 \tag{1}$$

$$M(NH_2NHCSNH_2)_2(CN)_2 + 14\ RNClNa\ (\text{or } 7\ RNCl_2) + 14\ H_2O \rightarrow M(CNO)_2 + 14\ RNH_2\ (\text{or } 7\ RNH_2) + 2\ CO_2 + 2N_2 + 14\ NaCl\ (\text{or } 14\,HCl) + H_2SO_4 + (NH_4)_2SO_4 \tag{2}$$

$$M(NH_2NHCSNH_2)_2(CNS)_2 + 20\ RNClNa\ (\text{or } 10\ RNCl_2) + 22\,H_2O \rightarrow M(CNO)_2 + 20\ RNH_2\ (\text{or } 10\ RNH_2) + 2\,CO_2 + 2\,N_3 + 20\,NaCl\ (\text{or } 20\,HCl) + H_2SO_4 + (NH_4)_2SO_4 \tag{3}$$

$$M(NH_2NCSNH_2)_2 + 12\ RNClNa\ (\text{or } 6\ RNCl_2) + 12\,H_2O \rightarrow MSO_4 + 12\,RNH_2\ (\text{or } 6\,RNH_2) + 2\,CO_2 + 2\,N_2 + 12\ NaCl\ (\text{or } 12\ HCl) + (NH_4)_2SO_4 \tag{4}$$

Where M = Pt or Pd; X = Cl, Br, I or NO_3.

Recommended Procedure

Add aliquots of the complex (2–30 mg) solution in pH4 buffer to 25 ml of 0·1 N CAT in an iodine flask. Shake the contents and set aside for about 30 minutes. Rinse down with about 20 ml of water, add 10 ml of 2N H_2SO_4 and 10 ml of 20% KI and titrate with 0·1 N sodium thiosulphate. Run a blank with CAT solution alone. A similar procedure is followed in oxidations with DCT[6].

Some typical results of analyses are given in Table I. The values are accurate with an errror around 0·5%.

It can be concluded from the investigations on metal complexes of TSC, that the number of ligand molecules present in a molecule of the complex, could easily be computed by oxidation with CAT or DCT.

TABLE I

Estimation of Platinum and Palladium complexes of thiosemicarbazide with chloramine–T and dichloramine–T

Complex	Range studied mg	% error in recovery		Complex	Range studied mg	% error in recovery	
		CAT	DCT			CAT	DCT
$Pt(TSC)_2Cl_2$	2–30	0·2–0·5	0·0–0·5	$Pd(TSC)_2Cl_2$	3–30	0·1–0·5	0·0–0·6
$Pt(TSC)_2Br_2$	3–46	0·2–0·3	0·1–0·3	$Pd(TSC)_2Br_2$	3–40	0·1–0·5	0·0–0·5
$Pt(TSC)_2I_2$	3–47	0·1–0·5	0·3–0·4	$Pd(TSC)_2I_2$	3–32	0·1–0·5	0·1–0·6
$Pt(TSC)_2(CN)_2$	2–33	0·0–0·4	0·0–0·4	$Pd(TSC)_2(CNS)_2$	2–30	0·3–0·5	0·0–0·5
$Pt(TSC)_2(CNS)_2$	2–31	0·0–0·5	0·0–0·5	$Pd(TSC)_2(NO_3)_2$	2–30	0·2–0·5	0·1–0·4
$Pt(TSC)_2(NO_3)_2$	2–24	0·2–0·6	0·3–0·6	$Pd(TSC\text{–}H)_2$	2–31	0·0–0·5	0·0–0·5
$Pt(TSC\text{–}H)_2$	2–31	0·2–0·5	0·1–0·5				

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1. Gaffre, A., In *Volumetric Analysis*, Vol. III, Ed. I. M. Kolthoff and R. Belcher, p. 386, Interscience, New York, 1957.
2. Pijck, J., Campe, A and Claeys, A., *Bull. Soc. Chim. Belges*, 1964, **73**, 898.
3. Suchomelova, L. and Zyka, J., *J. Electroanal. Chem.*, 1963, **5**, 57.
4. Mahadevappa, D. S. and Murthy, A. S. A., *Talanta*, 1970, **17**, 431.
5. — and —, *Curr. Sci.*, 1972, **41**, 100.
6. —, and Gowda, B. T., *Ibid.*, 1975, **44**, 306.
7. Morris, J. C., Salazar, J. R. and Wineman, M. A., *J. Amer. Chem. Soc.*, 1948, **70**, 2036.
8. Jacob, T. J. and Nair, C. G. R., *Talanta*, 1972, **19**, 347.
9. Jones, M. M., *Elementary Co-ordinatton Chemistry*, Prentice Hall, N.J., 1964, p. 254.
10. Chandrasekharan, M., Udupa, M. R. and Aravamudan, G., *Inorg. Chimica Acta*, 1973, **7**, 88.
11. Mahadevappa, D. S. and Murthy, A. S. A., *Australian J. Chem.*, 1972, **25**, 1565.
12. Suzuki, I., *Bull. Chem. Soc.*, Japan, 1962, **35**, 1286.
13. Higuchi, T., Ikeda, K. and Hussain, A., *J. Chem. Soc.*, B, 1967, p. 546.
14. Mahadevappa, D. S. and Gowda, B. T., *Talanta* (accepted for Publication).
15. Rao, V. R. S. and Murthy, A. R. V., *Curr. Sci.*, 1961, **30**, 176.

STUDY ON EQUILIBRIUM CONSTANTS OF UO_2^{2+} WITH MALONIC ACID AT DIFFERENT IONIC STRENGTHS

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ABSTRACT

The dissociation constants of the ligand and log K values of its complexes with uranyl ion have been investigated by Calvin-Bjerrum potentiometric titration technique at 30 ± 0·1° C and at various ionic strengths in aqueous medium. Uranyl forms both 1 : 1 and 1 : 2 complexes with malonic acid in the pH range 3–5. The plots of pK/log K *vs.* $\sqrt{\mu}$ were drawn to understand the exact complexation equlibria of 1 : 1 complex. The formation of 1 : 1 complex by the reaction $UO_2^{2+} + HL^- \rightleftharpoons UO_2L + H^+$ is confirmed. The thermodynamic constants at $\mu \rightarrow 0$ are reported.

INTRODUCTION

URANYL complexes of malonic acid were investigated by some workers[1,2]. Ramamoorthy and Santappa[3] have reported the uranyl complexes of malonic acid at 0·1 M $NaClO_4$. The present paper reports the study of the stability constants of UO_2^{2+} malonic acid system in aqueous medium at various ionic strengths.

EXPERIMENTAL

The details regarding the chemicals, apparatus are given in our earlier paper[4].

Calvin-Bjerrum Titration

The experimental procedure involved the potentiometric titration of carbonate free solution of (i) free $HClO_4$ ($4{\cdot}40 \times 10^{-3}$ M), (ii) free $HClO_4$ ($4{\cdot}40 \times 10^{-3}$ M) + malonic acid ($3{\cdot}02 \times 10^{-3}$ M), (iii) free $HClO_4$ ($4{\cdot}40 \times 10^{-3}$ M) + malonic acid ($3{\cdot}02 \times 10^{-3}$ M) + uranyl ion ($4{\cdot}32 \times 10^{-4}$ M) against sodium hydroxide (0·18 N) added from a microburette.

The ionic strength of the solution was maintained by the addition of appropriate amount of 1 M sodium perchlorate solution. The exact ionic strength of the solution was calculated by taking $\mu = \frac{1}{2} \Sigma\, c_i z_i^2$ where c_i and z_i are the concentration and valency respectively of *i*-th ion,

RESULTS AND DISCUSSION

The probability of complex formation between UO_2^{2+} and the ligand anion was assumed and the factors like hydrolysis of uranyl ion and the formation of polynuclear species were neglected on the following points.

(i) The pH of hydrolysis of uranyl ion, obtained from the deviation of uranyl ion curve from the acid veurc, was around pH 3·8. The departure of metal titration curve from the ligand curve was observed around pH 2·6. This indicated that the complexation occurs before the commencement of hydrolysis of ion. (ii) The solution being very dilute, the probability of existence of polynuclear species was negligible.

The proton-ligand formation number $\bar{n}_A$ and meta' ligand formation number $\bar{n}$ were calculated by Irving and Rossotti's expressions[5]. The approximate values of pK and log K were obtained by half integral method and accurate values were calculated by point-wise calculations[5]. The final values were also calculated by the method of least-squares.

The maximum value of $\bar{n}$ at each ionic strength was around 1·9. The metal-ligand ratio was taken as 1:5. No precipitation was observed in the metal complex titration upto pH 9. This showed that 1:1 and 1:2 complexes were formed before the commencement of hydrolysis. The thermodynamic values of pK_1, pK_2, $\log K_1$ and $\log K_2$ are given in Table I.

TABLE I

pK and log K thermodynamic values of UO_2^{2+}-malonic acid at various ionic strengths $t = 30°$ C

μ	pK_1	pK_2	$\log K_1$	$\log K_2$	$\log \beta$
0·032	2·83	5·59	5·87	3·54	9·41
0·052	2·80	5·50	5·75	3·66	9·41
0·072	2·73	·46	5·71	3·70	9·41
0·092	2·70	5·40	5·62	3·76	9·38
0·112	2·68	5·30	5·56	3·80	9·36

TABLE II

Slope values and thermodynamic constants

Equilibrium constants		$\triangle Z^2$	Thermodynamic contents at $\mu \to 0$
pK_1	..	2·17	3·00
pK_2	..	3·37	5·86
$\log K_1$	..	−3·87	6·20
$\log K_2$	..	+3·10	3·26

The stability constant data at different ionic strengths could be utilized to know the exact mechanism by which the complexation occurs at different pH. The data were analysed by Bronsted equation[6] as given

$$\log K = \log K° + A \triangle Z^2$$

where A is Debye-Huckel constant $(A = 0·5161)$[7], $\triangle Z^2$ is the difference in the square of the charges of product and reactant ion and K° is the formation constant at zero ionic strength.

The values of pK_1, pK_2, $\log K_1$ and $\log K_2$ when plotted against $\sqrt{\mu}$ gave the following results:

(i) All the points fell on a straight line in all the plots.

(ii) The observed slope values $(\triangle Z^2)$, along with expected, for various reactions are given as follows:

Reaction equilibrium	Constants	Slope values	
		Expected	Observed
$H_2L \rightleftharpoons HL^- + H^+$	pK_1	2	2·17
$HL^- \rightleftharpoons L^{2-} + H^+$	pK_2	4	3·37

The agreement between the observed and expected values of slope is fairly satisfactory.

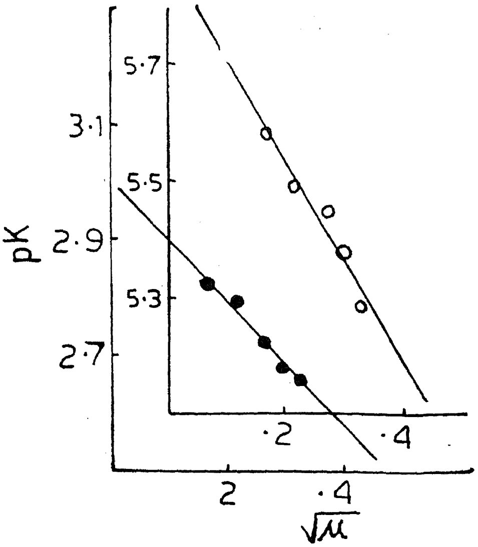

FIG. 1. Plots of pK_1 *vs.* $\sqrt{\mu}$ (●) and pK_2 *vs.* $\sqrt{\mu}$ (○) of the ligand malonic acid.

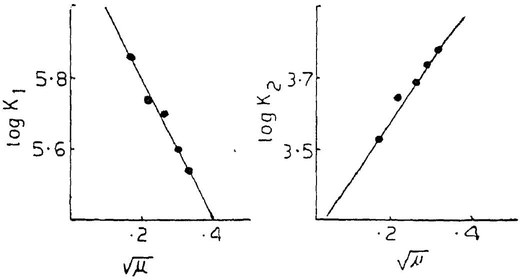

FIG. 2. Plots of $\log K_1$ *vs.* $\sqrt{\mu}$ and $\log K_2$ *vs* $\sqrt{\mu}$ for UO_2^{2+}-malonic acid complexes.

The complexation between UO_2^{2+} and malonic acid commences around pH 3. The dissociation of the first-COOH group of the acid occurs around the same pH ($pK \sim 2·83$). The ligand, therefore, may be present as a neutral malonic acid (H_2L) molecule or a malonate ion (HL^-). The alternate complexation equilibria for 1:1 complex are

$$UO_2^{2+} + H_2L \rightleftharpoons UO_2HL^+ + H^+$$

$$UO_2^{2+} + HL^- \rightleftharpoons UO_2L + H^+$$

The magnitude of $\triangle Z^2$ would be −2 or −4 for the first and the second equilibria respectively. The observed $\triangle Z^2$ (Table II) is in favour of second equilibrium.

The formation of 1:2 complex occurs around pH 5 at which the ligand is certainly present as HL^-. The 1:2 complexation therefore takes place by equilibrium

$$UO_2L + HL^- \rightleftharpoons UO_2L_2^{2-} + H^+$$

The observed ΔZ^2 confirm this speculation. The overall reaction is presented as

$$UO_2^{2+} + 2HL^- \rightleftharpoons UO_2L_2^{2-} + 2H^+$$

The changes in ionic strength could not affect this reaction. This has been concluded from the values ($\log \beta$) which remain constant.

ACKNOWLEDGEMENT

The authors wish to thank Prof. D. D. Khanolkar, Head of the Chemistry Department, Marathwada University, for his keen interest and encouragement in this work.

1. Athavale, V. T., Mahadevan, N., Mathur, P. K. and Sathe, R. M., *J. Inorg. Nucl. Chem.*, 1967, **29**, 1947.
2. Rajan, K. S. and Martell, A. E., *Ibid*, 1967, **29**, 523.
3. Ramamoorthy, S. and Santappa, M., *Bull. Chem. Soc. Japan*, 1969, **42**, 411.
4. Jahagirdar, D. V. and Khanolkar, D. D., *J. Inorg. Nucl. Chem.*, 1973, **35**, 921.
5. Irving, H. and Rossotti, H., *J. Chem. Soc.*, 1954, p. 2904.
6. Amis, A. S., *Solvent Effect on Reaction Rates and Mechanism*, Academic Press, N.Y., 1966, p. 24.
7. Robinson, R. A. and Stroke, R. H., *Electrolyte Solutiones*, 2nd Ed., Butterworths, 1959, p. 468.

NEUROMUSCULAR EFFECTS OF PHENETHYLBIGUANIDE (DBI)*

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ABSTRACT

Activity of Phenethylbiguanide (DBI, Phenformin) has been studied on neuromuscular junction. It is shown that, DBI inhibits neuromuscular junction. Experiments to elucidate mechanism of this inhibition have shown that DBI has membrane stabilizing activity.

INTRODUCTION

It is a long-known fact that the same structural features in different compounds are attended by similar pharmacological properties[1]. The structure of phenethylbiguanide (DBI, Phenformin), an oral hypoglycemic agent, consisting of a benzyl ring with two carbon atoms separating a biguanide radical, suggested the possibility of sympathetic action.

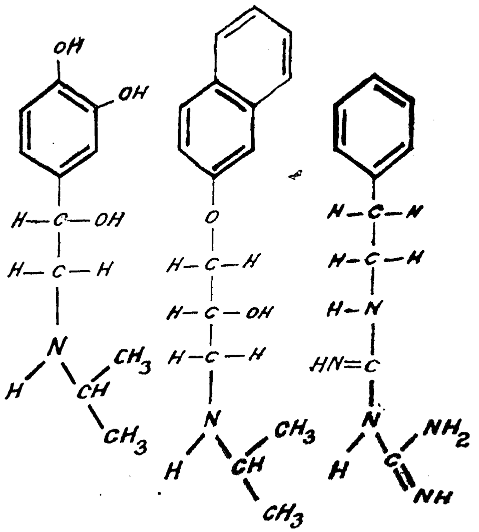
There are claims that DBI lacks a potentiating or blocking action, either to adrenaline or acetylcholine[2], and also that, only upon intravenous injection, in dogs, DBI produces an adrenergic block[3]. DBI is also reported to have negative chrontropic effect comparable to that of propranolol[4]. These contradictory reports, in spite of structural semblance of DBI with Beta-adrenergic drugs, suggested the need for critical reassessment.

There are reports that Beta-adrenergic drugs have both stimulatory[5] and inhibitory[6,7] activity at neuromuscular junction. Hence it was decided to study the activity of DBI on neuromuscular junction. Results obtained in such a study provide the basis for this report.

MATERIALS AND METHODS

(a) *Frog's Rectus Abdominis Muscle*

The procedure used is essentially as described by MacIntosh and Perry[8], with the following modifications. Magnification is ten fold, tension one gram; 20 to 30 min are allowed for relaxation in Frog Ringer solution (Composition, NaCl 0·65%, KCl 0·016%, $CaCl_2$ 0·012%, $NaHCO_3$ 0·01%, all W/V) before addition of physostigmine (Eserine) sulfate (2 mg/l) to sensitize the muscle. Another 20 to 30 min. are allowed for sensitization to be complete. Fifteen preparations were used for the study.



ISOPROTERENOL PROPRANOLOL DBI

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[*] Presented at the Second Southern Regional Conference of the Indian Pharmacological Society, Madras, October 30–31, 1974.

For acetylcholine, contractions are allowed to take place for 90 sec. The drug is then washed and the muscle allowed to relax. 90 sec. are allowed for D-Tubocurarine/DBI, action after which standard dose of acetylcholine is added to the same bath without washing, and the changes in response are recorded.

(*b*) *Frog's Gastrocnemius-Sciatic Muscle-Nerve Preparation*

Method used for mounting the preparation is the same as described by Harris[9]. For both direct and indirect stimulation of the preparation, square wave pulses of 4·0 volts and 1 per 10 sec. frequency were used. Total of twenty preparations were used in 10 of which end-plate block was established using D-Tubocurarine before application of DBI.

(*c*) *The Foot Withdrawal Reflex of the Frog*

The procedure adopted is the same as described by Perry[10]. Hydrochloric acid (0·1 N) was used as stimulus. Experiment was repeated in 12 preparations.

(*d*) *Surface Anaesthesia in Rabbits*

Surface anaesthesia in rabbits is tested as described by Weatherby[11]. A group of ten rabbits were used for the study.

RESULTS

(*a*) *Frog's Rectus Abdominis Muscle*:

DBI had a dose-proportionate inhibitory effect on acetylcholine induced contracture in all the preparations used. A period of 90 sec. of DBI contact with the preparation was sufficient to produce this inhibition. At least 20–30 min. waiting, with repeated washing was necessary to bring back the preparation to its pre-inhibitory responsive state (Fig. 1 *a*). By itself, DBI did not stimulate the preparation. A similarity of its action, as compared with D-Tubocurarine and Propranolol, can be seen in Fig. 1 *a*. Combination of DBI and D-Tubocurarine produced additive effect (Fig. 1 *b*).

(*b*) *Frog's Gastrocnemius-Sciatic Muscle-Nerve Preparation*

Local application of DBI on nerve decreased the response of indirect stimulation, when it was done, proximal to drug application. Response to indirect stimulation distal to drug application was unaltered (Fig. 2 *c*).

Injection of DBI (0·2 to 0·3 ml of 1% solution) to muscle belly produced a total block for both indirect and direct stimulations. This blockade was not reversed by Neostigmine.

In curarised preparations with end plate block, (*i.e.*, response to indirect stimulation absent and response to direct muscle stimulation present) injection of DBI to muscle belly abolished the response to direct stimulation also (Fig. 2 *d*).

(*c*) *The Foot Withdrawal Reflex of the Frog*

Absence of withdrawal reflex was seen after an average of 6–8 min. when 1% solution of DBI was used.

(*d*) *Surface Anaesthesia in Rabbits*

Effects of 1·0% solution of DBI after local application was observed for a period of 2½ hours.

DBI produced loss of corneal reflex and slight mydriasis. Light reflex was unaffected. Onset of loss of corneal reflex was on an average of 20–25 min. Corneal reflex did not reappear at the end of 2½ hours.

DISCUSSION

Acetylcholine response of frog's rectus abdominis muscle is blocked by DBI. Effects of D-Tubocurarine and DBI are synergistic.

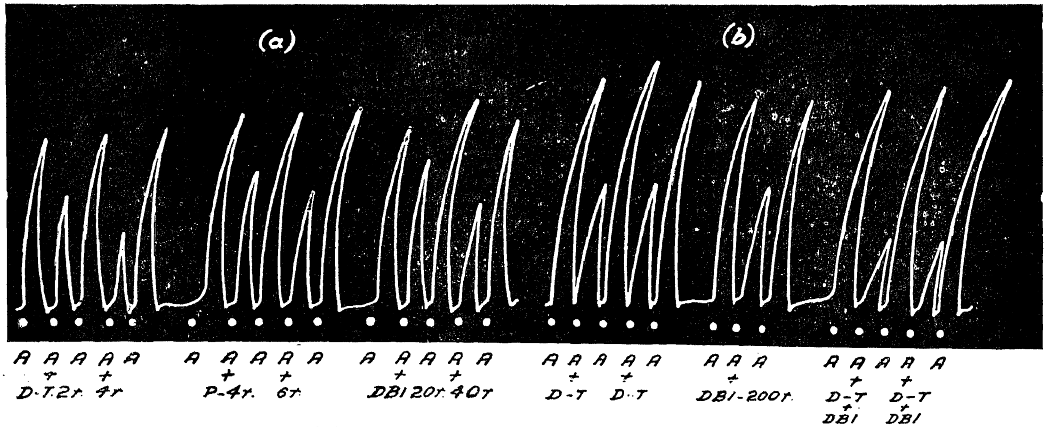

FIG. 1. Frog's rectus abdominis muscle preparation. (*a*) DBI showing inhibition of acetylcholine induced contracture. (*b*) Effect of combination of DBI and D-Tubocurarine on acetylcholine induced contracture. A, Acetylcholine 0·2 mcg. in (*a*) and 0·8 mcg. in (*b*); D–T, = D–Tubocurarine, 1·0 mcg, in (*b*); *p*, propranolol.

Responses to indirect and direct muscle stimulation in frog's gastrocnemius-sciatic muscle-nerve preparation are also blocked.

DBI has produced local anaesthetic action.

Mechanism of neuromuscular blocking action of DBI may be due to (1) Curare like—antidepolarising action. (2) Succinylcholine like—persistent depolarization action. (3) Membrane stabilization.

These similarities between Beta-adrenergic blocking drugs and DBI, warrant further study of DBI.

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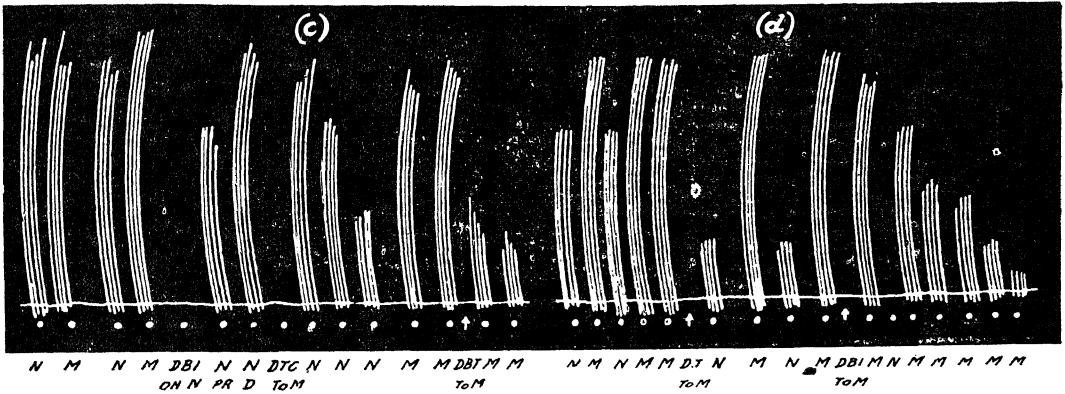

FIG. 2. Effect of DBI on frog's gastrocnemius—Sciatic muscle—nerve preparation. (*c*) DBI showing membrane stabilization at the point of application. (*d*) DBI Showing inhibition of direct muscle stimulation in curarised preparation. N, Nerve stimulation; M, Muscle stimulation; DBI, ON N, Application of DBI on nerve, D. TCTO M, Injection of D–Tubocurarine to muscle belly; DBI TO M, Injection of DBI to muscle belly; N PR, Nerve stimulation proximal to drug application; N D, Nerve Stimulation distal to drug application.

Unlike curare it also blocks direct muscle action and its action is not antagonized by neostigmine. Absence of initial contraction with DBI and abolition of response to direct stimulation of muscle precludes persistent depolarization mechanism.

The presence of local anaesthetic action with DBI and a similarity to propranolol action on neuromuscular junction stresses the possibility of a direct membrane stabilization action to DBI.

A comparison of DBI and propranolol is given in Table I. A number of points of similarity can be noted.

TABLE I

Some similarities between propranolol and DBI

Properties	Propranolol	DBI
1. Structural similarity	Present	Present
2. Hypoglycemia	Present[12]	Present[13]
3. Membrane stabilization		
(*a*) Skeletal muscle Membrane	Present[5]	Present**
(*b*) Cornea	Present[14, 15]	Present**
(*c*) Negative chronotropic effect	Present[16]	Present[14]

** — Finding of the present study.

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1. Alexander Gero, *Drill's Pharmacology in Medicine*, Ed. Dipalma, J. R., McGraw-Hill Book Company, New York, 1971, p. 82.
2. Ungar, G., Freedman, L. and Shapiro, S. L., *Proc. Soc. Exp. Biol. Med.*, 1957, **95**, 190.
3. Krantz, J. C. and Karr, C. J., *The Pharmacologic Principles of Medical Practice*, The Williams and Wilkins Company, Baltimore, 1969, p. 702.
4. Guruswami, M. N., Perumal, V. and Narayanan, T. K., *Ind. J. Physiol. Pharmacol*, 1971, **15**, 61.
5. Bowman, W. C. and Nott, M. W., *Pharmacol. Rev.*, 1969, **21**, 27.
6. Usubiaga, J. E., *Anaesthesiology*, 1968, **29**, 484.
7. Deshpande, V. R., Paranjpe, B. D. and Patki, V. P., *Indian J. Med. Res.*, 1974, **62**, 473.
8. MacIntosh, F. C. and Perry, W. M. L., *Methods in Medical Research*, Vol. 3, Year Book Publishers, Chicago, 1950, p. 78.
9. Harris, D. T., Gilding, H. P. and Smart, W. A. M., *Experimental Physiology*, J. and A. Churchill Ltd., London, 1956, p. 177.
10. Staff of the Department of Pharmacology, University of Edinburgh, *Pharmacological Experiments on Isolated Preparations*, E. and L. Livingstone, Edinburgh, 1970, p. 120.

11. Weatherby, J. H., *Evaluation of Drug Activities—Pharmacometrics*, Eds. Laurence, D. R. and Bacharach, A. L., Vol. 1, Academic Press, London, 1969, p. 206.
12. Harrison, D. C. and Griffin, J. R., *Circulation*, 1966, **34**, 218.
13. Travis, R. H. and Sayers, G., *The Pharmacological Basis of Therapeutics*, Ed. Goodman, L. S. and Gilman, A., The Macmillan Company, London, 1970, p. 1595.
14. Sinha, J. N., Jaju, B. P. and Misra, U. C., *Brit. J. Anaesth.*, 1967, **39**, 887.
15. Singh, K. P., Kulshrestha, O. P., Mahawar, M. M., Gautam, S. K. and Singh, M. M., *Lancet*, 1969, **2**, 158.
16. Gordon, K. M. and Abildskov, J. A., *The Pharmacological Basis of Therapeutics*, Eds. Goodman, L. S. and Gilman, A., The Macmillan Company, London, 1970, p. 723.

POLAROGRAPHIC STUDY OF MIXED LIGAND COMPLEXES : CADMIUM (II)-THIOUREA-FORMATE SYSTEM

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ABSTRACT

Cd (II) -thiourea-formate system was investigated polarographically in aqueous medium at 25 ± 0·5° C. The ionic strength was maintained at 2·0 M with potassium nitrate. Schaap and McMasters treatment of experimental data revealed the existence of [Cd (tu) (for)$_2$] and [Cd (tu)$_2$ (for)]$^+$ with their respective stability constants as log $\beta_{12} = 3\cdot65$ and log $\beta_{21} = 3\cdot99$. The negative value of β_{11} indicates the absence of [Cd (tu) (for)]$^+$ complex in solution.

INTRODUCTION

ALTHOUGH, most of the studies reported in literature have been made with the help of potentiometric[1] and spectrophotometric[1,2] techniques, the determination of the nature of mixed ligand complex formation by polarographic method has been receiving considerable attention in comparatively recent times[3]. This paper deals with the study of the composition and the stability constants of mixed ligand complex species which are formed by cadmium (II) with formate and thiourea. The simple systems, namely, Cd (II)-formate[4,5] and Cd (II)-thiourea[6,0] were previously studied polarographically at different ionic strengths and in aqueous and non-aqueous media. However, these simple systems were reinvestigated in the present work in order to obtain stability constants of complexes under identical conditions maintained for mixed systems.

EXPERIMENTAL

Potassium nitrate was used as a supporting electrolyte to maintain the ionic strength at 2·0 M in all cases. Triton X-100 (0·004%) was used as a maximum suppressor. The pH of the solutions to be polarographed was in the range of 6·6 to 6·8. A Cambridge Pen Writing Polarograph was used to record the polarograms. Purified nitrogen was bubbled to exclude the dissolved oxygen. All half-wave potentials refer to saturated calomel electrode (S.C.E.).

The dropping mercury electrode had the following characteristics:

$t = 3\cdot0$ sec in $0\cdot1$M KNO$_3$ (open circuit)

$m = 1\cdot97$ mg sec^{-1}, $m^{2/3} \times t^{1/6} = 1\cdot88$ mg$^{2/3}$ sec$^{-1/2}$

The measurements were carried out in an air conditioned room where the temperature was maintained at 25 ± 0·5° C.

RESULTS AND DISCUSSION

The Stability Constants of Cd (II)-Thiourea System:

All the solutions contained 1mM Cd (II) and 0·004% Triton X-100. The concentrations of thiourea varied from 0·05 to 0·6 M. The half-wave potential of Cd (II) (2·0 M KNO$_3$) was $-0\cdot593$ V which shifted to more negative potential by the addition of thiourea showing the complex formation.

The relationship between $-(E_{\frac{1}{2}})_c$ and log C_L, where C_L is the concentration of thiourea (tu), was not linear but gave a smooth curve with three segments indicating the existence of 1 : 1, 1 : 2 and 1 : 3 complex species. The method of DeFord and Hume[10] was, therefore, applied to evaluate the stability constants which were found to be as under:

$\log\beta_{10} = 1\cdot69 \pm 0\cdot11$, $\log\beta_{20} = 1\cdot95 \pm 0\cdot10$ and $\log\beta_{30} = 3\cdot47 \pm 0\cdot03$. Our values are nearly identical to those observed by Lane *et al.*[6] and Migal *et al.*[7,8]. The slight divergence may be due to the differences in the ionic strengths and the media used.

The Stability Constants of Cd(II)*-Formate System*:

The concentration of potassium formate was varied from 0·1 to 1·3M. Three complex species 1:1, 1:2 and 1:3 were detected and their stability constants were $\log\beta_{01} = 0\cdot95 \pm 0\cdot10$, $\log\beta_{02} = 0\cdot60 \pm 0\cdot11$ and $\log\beta_{03} = 1\cdot55 \pm 0\cdot04$ which followed the order $\beta_{01} > \beta_{02} > \beta_{03}$. Gaur *et al.*[4], have obtained a similar trend in β values at 1·0 M (KNO_3) ionic strength. Our values do not agree satisfactorily with those obtained by Hershenson *et al.*[5] at 2·0M ionic strength maintained by sodium perchlorate.

The Cd (II)-Thiourea-Formate System:

Polarographic measurements of mixed ligand system were carried out under such conditions that the concentration of one ligand was held constant while that of the other was varied. A study of the present mixed ligand system was investigated in two sets of experiments at two constant concentrations of formate, *i.e.*, at 0·30 and 0·60 M. These two concentrations of formate were chosen in such a way that 1:1 and 1:2 Cd (II)-formate complexes were in predominant quantities at these concentrations. Thiourea concentration was varied from 0·05 to 0·60 M in each case. For the first set, solutions contained 1mM Cd (II), 0·004% Triton X-100, 0·30 M formate and thiourea concentration was varied as stated above. The same procedure was repeated in the case of second set in which the formate concentration was kept constant at 0·60M.

The polarograms for the solutions were well defined. The plots of E.d.e. *vs* log $r/i_d - i$ were linear with a slope of 30 ± 4 mV.

The values of $-(E_{\frac{1}{2}})$ and F_{ij} functions at both the concentrations of formate are set out in Tables I and II.

In both the sets, the shift in half-wave potential of Cd (II) to more negative potential with an increase in the thiourea concentration was observed. The fact that these shifts are greater in magnitude as compared with those obtained in the absence of formate clearly shows the formation of mixed ligand complexes.

The plots of $-(E_{\frac{1}{2}})_c$ *vs* log C_L (Fig. 1) for both the concentrations of formate are smooth curves with three segments. The slopes of the segments indicate the presence of three mixed igand complexes. The slopes of these segments for formate concentration of 0·30 M, show the addition of (i) one thiourea molecule in the range of 0·05 to 0·10 M, (ii) two molecules in the range of 0·15 to 0·40 M and (iii) three molecules above the concentration of 0·40 M of thiourea. Similarly, the corresponding concentration range values for 0·60 M formate are respectively,

TABLE I
F_{10} as function of (tu)

Cd (II) = 1mM
[for] = 0·3M

Temp. 25 ± 0·5° C

$\mu = 2\cdot0M\ (KNO_3)$
$-(E_{\frac{1}{2}})_s = 0\cdot593$ V

Concentration of ligand (tu) M	$-E_{\frac{1}{2}}$ *vs* S.C.E. at [for] 0·00 M	at [for] 0·30 M	$\log I_M/I_C$	Slope (mV)	$F_{00} \times 10^{-2}$	$F_{10} \times 10^{-2}$	$F_{20} \times 10^{-2}$	$F_{30} \times 10^{-3}$
0·05	0·606	0·634	0·0437	31·0	0·275	1·504	..	..
0·10	0·618	0·643	0·0531	30·0	0·595	3·955	..	..
0·15	0·630	0·653	0·0626	30·5	1·280	7·266	40·40	..
0·20	0·636	0·662	0·0626	30·0	2·700	12·540	56·70	13·350
0·25	0·640	0·667	0·0626	30·0	4·020	15·280	56·32	10·528
0·30	0·649	0·673	0·0626	30·3	6·205	20·001	62·70	10·900
0·40	0·660	0·680	0·0927	30·5	12·130	29·825	71·52	10·380
0·50	0·670	0·688	0·0927	30·5	21·750	43·100	83·80	10·730
0·60	0·675	0·697	0·0927	30·3	44·270	73·450	120·00	15·060

log A = 1·30, log B = 2·08±0·07, log C = 3·48±0·15 and log D = 4·01±0·04.
[For] = Formate, $F_{10} = F_{10}$ [tu, for].

TABLE II

F_{10} as function of (tu)

Cd (II) = 1mM
[for] = 0·6M

Temp. 25 ± 0·5° C

$\mu = 2 \cdot 0M\ (KNO_3)$
$-(E_{\frac{1}{2}})_s = 0 \cdot 593\ V$

Concentration of ligand (tu) M	$-E_{\frac{1}{2}}$ vs S.C.E. at [for] 0·00 M	$-E_{\frac{1}{2}}$ vs S.C.E. at [for] 0·60 M	$\log I_M/I_C$	Slope (mV)	$F_{00} \times 10^{-2}$	$F_{10} \times 10^{-2}$	$F_{20} \times 10^{-3}$	$F_{30} \times 10^{-4}$
0·05	0·606	0·650	0·0626	30·0	1·058	11·160	..	..
0·10	0·618	0·660	0·0833	30·3	2·326	18·260	8·260	..
0·15	0·630	0·667	0·0723	30·2	4·025	23·500	9·000	2·666
0·20	0·636	0·676	0·0723	30·0	6·500	30·000	10·900	2·500
0·25	0·640	0·678	0·0927	30·4	10·430	39·720	11·000	3·955
0·30	0·649	0·684	0·0723	29·6	16·100	52·000	14·000	3·000
0·40	0·660	0·693	0·0927	30·5	34·100	84·000	18·500	4·125
0·50	0·670	0·700	0·0927	30·5	58·000	115·000	21·000	3·200
0·60	0·675	0·706	0·0927	30·1	92·900	154·000	24·000	3·166

log A = 1·69, log B = 3·0 ± 0·09, log C = 3·69 ± 0·08 and log D = 4·48 ± 0·08.
[For] = Formate, $F_{10} = F_{10}$ [tu, for].

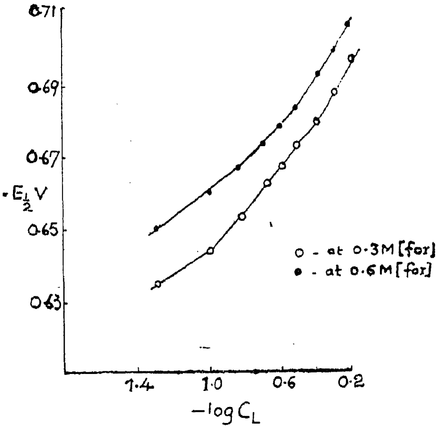

FIG. 1

(i) 0·05 M to 0·20 M, (ii) 0·25 to 0·40 M and (iii) above 0·40 M.

The method of Shaap and McMasters[3] was used to determine the following values of A, B, C and D at both the concentrations of formate. At 0·30 M formate concentration: log A = 1·30, log B = 2·08 ± ± 0·07, log C = 3·48 ± 0·15 and log D = 4·01 ± 0·4 and at 0·60 M formate concentration: log A = 1·69, log B = 3·0 ± 0·09, log C = 3·69 ± 0·08 and log D = 4·48 ± 0·08. From these constants, the following values of stability constants of mixed ligand complexes were calculated: $\log \beta_{12} = 3 \cdot 65$, and $\log \beta_{21} = 3 \cdot 99$ (mean value). The negative value of β_{11} indicated the absence of the $[Cd\,(tu)\,(for)]^+$ complex in the solution. Log D values in the two sets are 4·01 ± 0·04 and 4·48 ± 0·08 and are higher than $\log \beta_{30}$, the stability constant of the simple $[Cd\,(tu)_3]^{2+}$ complex. This higher value of log D may probably be due to the incomplete dissociation of $[Cd\,(tu)_2\,(for)]^+$ complex in aqueous medium.

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1. Martell, A. E., *Stability Constants*, Special Publication No. 17. and Supplement No. 1, 1971, Tha Chemical Society, London.

2. Tanaka, M., *J. Inorg. Nucl. Chem.*, 1974, **36**, 151.
3. Schaap, W. B. and McMasters, D. L., *J. Amer. Chem Soc.*, 1961, **83**, 4699.
4. Gaur, J. N. and Jain, D. S., *Australian J. Chem. Soc.*, 1965, **18**, 1687.
5. Harshenson, H. M., Brooks, R. T. and Murphy, M. E., *J. Amer. Chem. Soc.*, 1957, **79**, 2046.
6. Lane, T. J., Ryan, J. A. and Britten, E. F., *J. Amer. Chem. Soc.*, 1958, **80**, 315.
7. Migal, P. P. K. and Tsiplyakova, V. A., *Zh. Neorgan. Khim.*, 1964, **9** (3), 601.
8. Migal, P. K and Tsiplyakova, V. A., *Zh. Neogran. Khim.*, 1964, **8**, 629.
9. Dhuley, D. G., *Ph.D. Thesis*, Marathwada University, Arurangabad, India, 1973.
10. DeFord, D. and Hume, D. N., *J. Amer. Chem. Soc.*, 1951, **73**. 5321.

EVOLUTION OF FOLIAR VENATION IN EMBRYOBIONTA

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Abstract

A survey has been carried out of the foliar venation in the various 'type' taxa of the Embryobionta as suggested in the classification proposed by Cronquist, Takhtajan and Zimmermann (1966). The study includes not only the true leaves of higher cryptogams but also the scaly leaves of *Psilotum, Rhynia, Equisetum,* and *Ephedra* have been taken into account. It is seen that usually there is a gradation of complexity in this feature as we go higher up in the ladder of evolution. In several representatives, however, the veins have become less numerous and rarified.

Introduction

PHYLOGENY is generally regarded as the evolutionary history of a taxon or a group of organisms and attempts to account for its origin and development. As such one of the prerequisite of the phylogenetic studies in Botany is the determination of the origin and relationships of all taxa of both extinct and present-day plants and the classification or 'grouping' of them according to a system that will indicate their genetic or 'blood' relationships. Sufficient work is available on this aspect now taking into account various features or attributes, both of the vegetative and reproductive parts, like archegonium (or the female gametophyte as a whole), pollen (or the male gametophyte as a whole), etc. Similarly, floral characters, nodal anatomy, foliar anatomy, cytology, biometrical genetics, chemotaxonomy, and numerical taxonomy are regarded important parameters, worthy of consideration, from this point of view.

The term 'embryobionta' has been coined by three of the leading present-day taxonomists—Arthur Cronquist (New York), Armen Takhtajan (Leningrad), and Walter Zimmermann (Tübingen) in the year 1966. They also propounded a new system of classification for the embryo-bearing plants. Accordingly, they placed them under the sub-kingdom 'Embryobionta' which includes plants ranging from Bryophyta to Phanerogams of the older systems of classification. They also advanced concrete arguments in support of this new approach.

Necessity was felt by us to provide an understanding of the various stages and aspects of evolution of the foliar features into different groups of the embryobionta based on this arrangement. The present communication is restricted to a consideration of the foliar vascularization alone whereas the epidermal characters form the subject-matter of another article to be published shortly.

Experimental

Sources of information.—It must be admitted at the outset that in carrying out this survey, we encountered difficulties in procuring both the fossil as well as living materials. Howsoever we wished to study the fossils in detail, we could not do so because of the insufficient information and preparations at our disposal. Specimens of the living materials were collected locally or from the hill stations. Specimens from the herbarium sheets of the Delhi University Botany Department and those from the museum also served us very well. We also attempted to procure data about the fossils from leading palaeobotanists in India and abroad by correspondence but got limited success.

Preparation of the material for microscopic study.—Following the methodology proposed by Paliwal and Kakkar (1969), dried leaves were at first kept in hot water for half an hour, transferred to 3% NaOH for overnight, washed thoroughly with water the next day and placed in the chloral hydrate (100%) + hydrogen peroxide mixture for three hours to overnight. This was later passed through a chloral hydrate series (75%, 50%, 25%) keeping in each grade for 10 minutes and washed thoroughly with distilled water. It was dehydrated through an ascending series of alcoholic grades and stained with safranin dissolved in a mixture of absolute alcohol and xylol (1:1) for 10 minutes. Mounting was done in piccolyte.

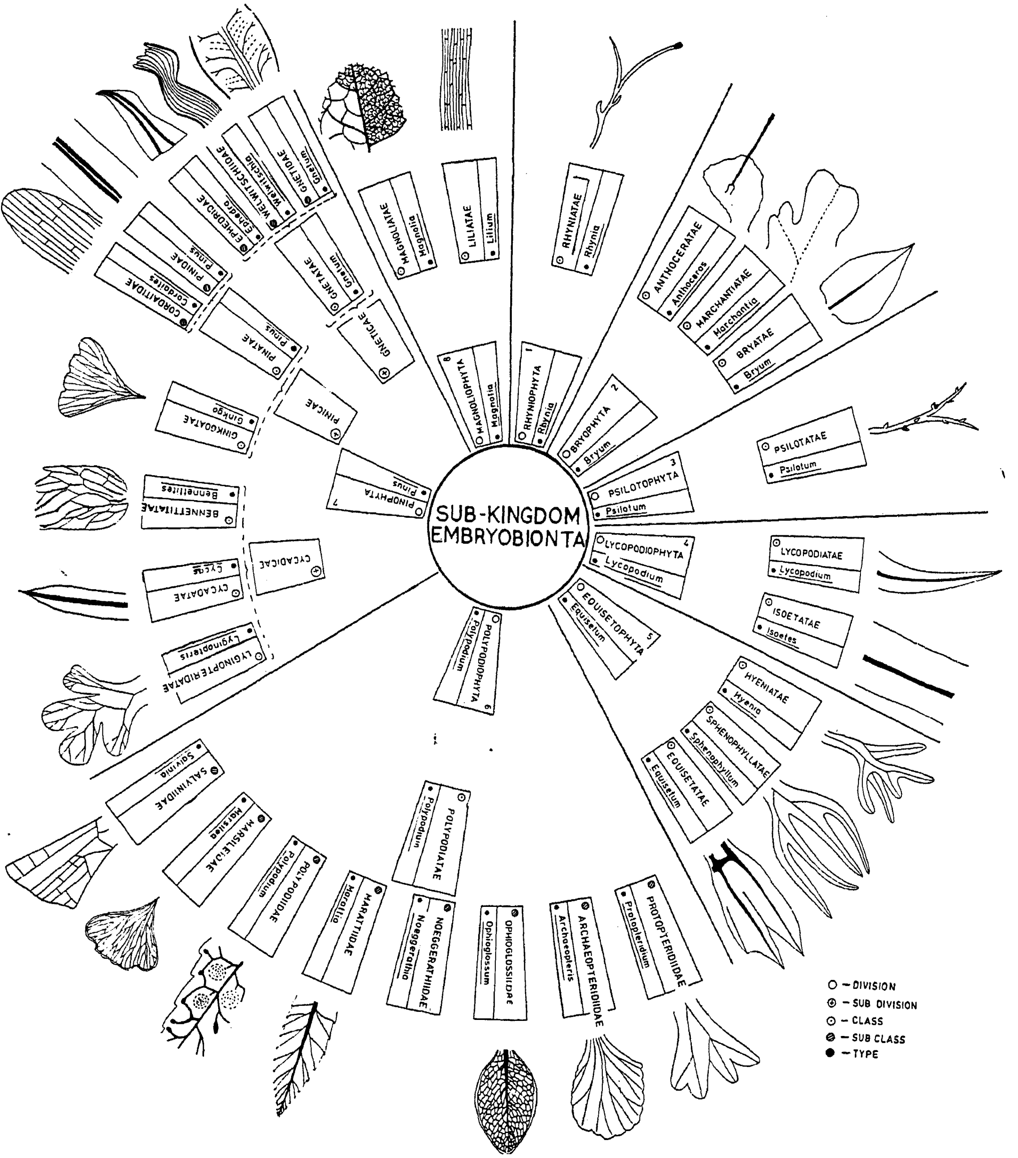

FIG. 1. Foliar Venation in vascular plants.

RESULTS

Morphology of the Leaves and Gross Venation.— In *Rhynia* species and *Psilotum nudum* true leaves are not present but only scaly leaves occur without any vasculature. In *Marchantia polymorpha* the 'midrib' is quite prominent which dichotomizes along with the thallus. However, no such inner tissue differentiation is noticeable as to warrant its description as a vein. In *Anthoceros himalayensis*, the midrib is totally absent. The leaves are leptophyll* in *Bryum* sp., *Psilotum nudum*, and *Lycopodium selago*; nanophyll in *Isoetes coromandelina*, *Marsilea minuta*, *Salvinia natans*, *Pinus roxburghii* and *Ephedra foliata*; microphyll in *Ophioglossum reticulatum*, *Cycas circinalis*, *Ginkgo biloba* whereas in *Marattia* sp., *Polypodium spinidosum*, *Gnetum*

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* Terminology after Dilcher (1974).

gnemon, and *Magnolia stellata* they are of the mesophyll type.

Origin of Variability.—Primitive vascular plants like *Lycopodium* and *Equisetum* have a primary or 1° vein while those like *Rhynia* and *Psilotum* which are at the bottom of this category lack it completely. Nodal anatomy in all these representatives such as the species of *Lycopodium*, *Psilotum* and *Selaginella*, etc., is alacunar meaning thereby that the vascular system of the leaf does not arise from the main stele (or the stem stele). This may form a part of such a discussion where it is opined that the leaf traces form the stem stele and the stem does not have any vasculature of its own. In *Marchantia polymorpha* the 'midrib' appears distinct externally but it is devoid of any vasculature altogether whereas in *Bryum* sp. and *Cycas circinalis* also a single or 1° vein alone is present. An identical or univeined condition is seen in the young leaves of *Santalum album* (Aggarwal *et al.*, unpublished information) and *Cercis siliquastrum* (Slade, 1957) at the ontogenetic stages of the leaves. This indicates that ontogeny repeats phylogeny.

Primary or secondary vein systems are present in *Marattia* sp., *Polypodium* sp., *Marsilea minuta*, *Salvinia natans* and *Ginkgo biloba*; a little more advanced or 1°, 2° and 3° vein system is present in *Ophioglossum reticulatum*, *Gnetum gnemon*, *Magnolia stellata*, and *Lilium giganteum*. This may be due to increased independence of these taxa from the hydrophytic habitat.

Steps of Evolution.—In taxa like *Bryum* sp., *Lycopodium selago*, *Equisetum ramosissimum*, *Cycas circinalis*, *Pinus roxburghii*, and *Ephedra foliata* only primary or 1° veins are present. However, 1° and 2° vein system is present in *Marattia* sp., *Polypodium spinidosum*, *Marsilea minuta*, *Salvinia natans*, and *Ginkgo biloba* whereas *Ophioglossum reticulatum*, *Gnetum gnemon* and *Magnolia stellata* leaves possess 1°, 2° and 3° vein systems. The most complex vasculature among the taxa studied is of course seen in *Magnolia stellata* wherein the leaves exhibit a further differentiation of the venation system (beyond 3°). Taking stock of the branching of the lateral veins, we find that the primary or 1° vein system has a single midvein which remains unbranched in *Bryum* sp., *Lycopodium selago*, *Isoetes coromandelina*, *Equisetum ramosissimum*, *Cycas circinalis*, *Pinus roxburghii* (where two primary or 1° veins are present) and *Ephedra foliata*. Secondary or 2° vein system has been recorded in *Ophioglossum reticulatum* and *Marsilea minuta* (which also possesses an intramarginal vein). Dichotomously branched situation is seen in *Marattia* sp., *Ginkgo biloba* and *Magnolia stellata* forming compartments or areoles. Distinct areole formation also occurs in *Ophioglossum reticulatum*, *Polypodium spinidosum*, *Marsilea minuta*, *Salvinia natans*, *Gnetum gnemon*, *Magnolia stellata*, and *Lilium* sp.

The Fossil Forms.—*Rhynia* lacks 1° or primary vein system in the fossils. In taxa like *Hyenia* sp., *Sphenophyllum* sp., *Protopteridium* sp, *Archaeopteris* sp., *Bennettites* sp. only primary or 1° veins are present Primary or 1° single and dichotomously branched veins are seen in *Hyenia* sp., *Sphenophyllum* sp., and *Protopteridium* sp. The primary or 1° veins are several and branched in *Archaeopteris* sp., *Bennettites* sp., and *Cordaites* sp. Arcole formation also seems to occur in the leaves of *Bennettites* sp.

GENERAL COMMENTS AND CONCLUSIONS

Taxonomic evidence (or characters) can be drawn from any part and phase of development of a plant. In practice, however, overwhelming reliance is placed on morphological features, at least for grouping of the various representatives and thereupon erecting classifications. It is also a truthful realization that the primitive morphology of a given part is usually accompanied by specialization of other structures which Bower (1923) called as 'phyletic inertia'. In this connection it is interesting to note that Bailey (1957) has commented on the futile time and effort put in by many workers to show or even 'prove' that certain organs or features of the plant are inherently more conservative than others and, therefore, should have a more dominant role in the analysis of classification and phylogeny. As such the utility of as many characters as can be studied becomes apparent. Taking clue from this fact the argument put forth by some anatomists that endomorphic features are more important than exomorphic structures becomes completely ill founded. Analysis of the angiosperms as a whole shows that such an assumption cannot be fully justified and that each morphological or histological attribute is fundamentally similar and potential of being reckoned for analysis in systematics. It remains, however, true that when looked at under the microscope or with the naked eye, some features are found to be relatively stable in certain groups of plants and show considerable variations in others. It is therefore evident that endomorphic characters such as anatomy are neither more nor less reliable than exomorphic features; perhaps they are of equal valid use. They give us, as Bailey says, more string to our bow.

During more recent years, leaf anatomical features such as stomata, trichomes, venation, etc., have been recognized to play an increasingly important role in the formulation of natural groups. They are also widely used in other aspects of taxonomy and have been largely applied to the elucidation of 'phylogenetic' relationships.

The present observations reveal that there is no linear evolution even among the taxa grouped together indicating that these must have originated from different and perhaps also 'distant' ancestors. Variable opinions have been expressed in the literature from time to time with regard to the probable lineage of the vascular plants *vis-a-vis* the foliar venation. For instance, Zimmermann (1959) thinks that in gymnosperms the dichotomous foliar venation is the retention of a truly primitive type of vasculature. Hammen (1948) and Lam (1959) consider that instances of primitive dichotomy are still present in the forked pattern of branching of the major leaf veins. Hammen also adds that the accompanying network of veinlets tends to darken the picture.

Looking at the information available for Magnoliophyta we find that Foster (1959 *a*) recorded for *Kingdonia uniflora* leaf an open type of dichotomous venation pattern which he described to be strikingly like the venation of the leaves of *Ginkgo biloba* and certain fern genera. Other authentic examples of open dichotomous venation in the angiosperms are known in *Circaester* (Foster, 1961 *b*; 1963) and *Utricularia* (Subramanyam, 1969; *see also* Subramanyam and Banerjee, 1967). Foster (1959 *b*) has discussed the question whether the reticulate type of venation so characteristic of living angiosperms, may not have evolved from open dichotomous vasculature. According to Hutchinson (1959), Foster (1959 *a*), and Foster and Arnott (1960), this possibility is clearly indicated by the venation pattern in *Kingdonia uniflora*. In this genus the vascular supply at the nodal level consists of four collateral traces which diverge from a single gap into a sheathing leaf base. According to Foster and Arnott (1960) this type of nodal anatomy is comparable to the unilacunar, 2- and 4-traced system found in the cotyledonary nodes of some of the woody Ranales. The vascular system of the tepals is also 2-traced and of the unilacunar type (Foster, 1961 *a*). Foster and Arnott (1960) consider that from a morphological point of view *Kingdonia* type of venation is comparable to *Sphenophyllum*, some ferns and *Ginkgo*. This is perhaps an indication of the polyphyletic origin of the different groups of plants included in the subkingdom Embryobionta, although at places we find that some idea becomes available of the 'slide' of venation of representatives of one type becoming like that of the other.

ACKNOWLEDGEMENTS

The authors are extremely grateful to their colleagues at the laboratory for their kind co-operation and valuable suggestions.

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1. Bailey, I. W., *J. Arnold Arbor.*, 1957, **38**, 243.
2. Bower, F. O., *The Ferns (Filicales)*. Cambridge, 1923-28.
3. Cornquist, A., Takhtajan, A. and Zimmermann, W., *Taxon*, 1966, **15**, 129.
4. Dilcher, D. L., *Bot. Rev.*, 1974, **40**, 1.
5. Foster, A. S., *Notes Roy. Bot. Gard. Edinb.*, 1959 *a*, **23**, 1.
6. —, *Proc. IX Internat. Bot. Congr.*, 1959 *b*, **2**, 119.
7. —, *J. Arnold Arbor.*, 1961*a*, **42**, 397.
8. —, In *Recent Advances in Botany*. Ed. R. D. Preston, Univ. of Toronto Press, 1961*b*, p. 971.
9. —, *J. Arnold Arbor.*, 1963, **44**, 299.
10. — and Arnott, H. J., *Amer. J. Bot.*, 1960, **25** 684.
11. Hammen, van der, *Blumea*, 1948, **6**, 290.
12. Hutchinson, J., *Families of the Flowering Plants*, Oxford Univ Press, 1959.
13. Lam, H. J., *Trans. Bot. Soc.*, Edinburgh, 1959, **38**, 100.
14. Paliwal, G. S. and Kakkar, *Lalita, Acta Agronom. Acad. Scien. Hungar.*, 1969, **18**, 406.
15. Slade, B. F., *New Phytol.*, 1957, **56**, 281.
16. Subramanyam, K., *Proc. XI Internat. Bot. Congr. Seattle*, 1969, p. 212.
17. — and Banerji, L. K., *Proc. 54th Indian Sci. Congr.* (Varanasi), Pt. III: *Abstr.*, 1967, p. 315.
18. Zimmermann, W., *Die Phylogenie der pflangen*, ed. 2, Guotar Fischer Verlag, Stnttgart, 1959.

THE NATIONAL SYMPOSIUM ON RADIATION PHYSICS

The National Symposium on Radiation Physics sponsored by the Department of Physics, University of Mysore, Mysore, will be held during June 10-12, 1976 at the University Physics Department. The last date for receiving abstracts–April 15, 1976, papers–May 30, 1976.

Details can be had from Prof. B. Sanjeevaiah, Convener, National Symposium on Radiation Physics (1976), Department of Physics, University of Mysore, Manasagangotri, Mysore 570 006.

SEMINAR ON ENVIRONMENTAL GEOSCIENCE (JUNE 26-28, 1976)

The Indian Institute of Geological Education has proposed to conduct, in collaboration with other scientific societies, a Seminar on the subject mentioned above along with a workshop on the same theme in June 1976. Further details can be had from the Secretary, Prof. S. Sambegowda, Central College, Bangalore 560 001.

LETTERS TO THE EDITOR

MEASUREMENT OF K-SHELL PHOTOELECTRIC CROSS-SECTIONS FOR SOME CHARACTERISTIC X-RAYS

BECAUSE of the non-availability of clean and strong low energy photon sources, external conversion X-rays in various suitable targets have been produced and successfully used to measure relative K shell photoelectric cross-sections in Y for incident energies 17·781, 22·581, 23·618, 25·770, 29·208, 35·478, 41·006 and 43·949 keV in a double reflection geometry experiment as shown in Fig. 1.

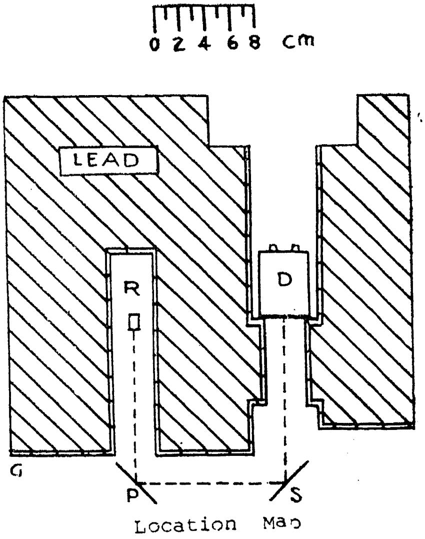

FIG. 1. Experimental arrangement for the double reflection experiment for the measurement of photoelectric cross-sections; R, Radiation source Tm^{170}; P, Primary target; S; Secondary target; D, Detector. G, Graded absorber of Sn, Cu and Al.

A brief outline of the procedure of measurement and the results obtained are given in this communication. The details will be published elsewhere.

A Y target placed at S was irradiated, turn, with characteristic X-ray of elements, Mo, Ag, Cd, Sn, I, Ce, Sm and Gd, obtained as a result of interaction of photons from a ~ 2 Curie Tm^{170} source R with their respective targets placed at P. The intensity of the resulting K X-rays in Y was measured with a thin NaI (Tl) detector placed at D. The intensities of the K X-rays of various target elements, used as primary targets, falling on Y target were measured with a calibrated NaI (Tl) detector placed at S. Background due to the scattering of photons was taken care of by subtracting the spectrum recorded with an equivalent aluminium target from the main spectrum in each case.

The ratio of the K shell photoelectric cross-sections for the primary energies E_1 and E_2 corresponding to primary targets P_1 and P_2 is given as

$$\frac{\sigma(E_1)}{\sigma(B_2)} = \frac{N(P_2)}{N(P_1)} \cdot \frac{Ny(P_1)}{Ny(P_2)} \cdot \frac{\beta(E_2)\ \epsilon(E_1)}{\beta(E_1)\ \epsilon(E_2)} \qquad (1)$$

where $N(P)_2/N(P_1)$ is the ratio of the counting rates under photopeaks of primary K X-ray spectra taken with the counter placed at S for primary targets P_2 and P_1. $N_Y(P_1)/N(P_2)$ is the ratio of the counting rates under photopeaks of spectra when Y target is irradiated with primary X-rays from targets P_1 and P_2 respectively. $\beta(E_2)/\beta(E_1)$ and $\epsilon(E_1)/\epsilon(E_2)$ are the ratios of the self-absorption corrections for Y target and effective efficiencies of the counter used for the detection of primary X-rays, at energies E_1 and E_2 respectively.

Measuring $N(P_2)/N(P_1)$ and $N_Y(P_1)/N_Y(P_2)$ from the respective spectra and calculating $\beta(E_2)/\beta(E_1)$ and $\epsilon(E_1)/\epsilon(E_2)$ as described earlier[1,2] $\sigma(E_1)/\sigma(E_2)$ were determined and are compared with theoretical calculations of Scofield[3] in Table I.

TABLE I

The present measurements of the relative K shell photoelectric cross-sections of Y are compared with the theoretical calculations of Scofield[3]

Primary target element	Average K X-ray energy (E_2) in keV	$\sigma(E_1{}^*)/\sigma(E_2)$	
		Present measurements	Theoretical calculations[5]
Mo	17·781	1·000 ±0·033	1·000
Ag	22·581	1·887 ±0·066	1·833
Cd	23·618	2·220 ±0·078	2·115
Sn	25·770	2·671 ±0·093	2·619
I	29·208	3·822 ±0·134	3·606
Ce	35·478	5·875 ±0·235	5·789
Sm	41·006	8·630 ±0·345	8·627
Gd	43·949	10·865 ±0·436	10·476

$E_1{}^*$ is taken as 17·781 keV.

The experimental results show good agreement with theory. It may be noted that the errors involved in the present measurements are less than 5% as compared to 10% in the earlier measurements made by using internal conversion X-rays[4,5]. To the best of our knowledge, the data reported above are new and no other measurements are yet available in this energy region.

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Patiala 147 002, *July* 11, 1975.

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1. Shute, G. G. and Sood, B. S., *Proc. Roy. Soc.* (London), 1960, 257 A, 52.
2. Singh, M., Anand, S. and Sood, B. S., *Indian J. Pure Appl. Phys.*, 1963, 1, 305.
3. Scofield, J. H., *Lawrence Livermore Laboratory*, report UCRL-51326, 1973 (unpublished).
4. Allawadhi, K. L., Mohanjit Singh, and Sood, B. S., *Curr. Sci.*, (India), 1973, 42, 86.
5. — and Sood, B. S., *Phys. Rev.*, 1975, 11, 1928.

EFFECT OF SOLVENT ON POLARIZATION OF LUMINESCENCE

IT is known that the polarization of luminiscence of solution changes due to several factors, *viz.*, viscosity of the solvent[1], migration of the excited state from the originally excited molecule to a neighbouring molecule[2], shape and size of the fluorescent molecule[3], etc. The present report deals with the effect of dielectric constant/dipole moment of the solvent as an extrinsic cause of depolarization. The polarization of fluorescein in different solvents (con. ~ 10^{-5} g/cc) has been measured at room temperature (~ 20° C) with an Aminco Bowman spectrophotofluorometer. The solvents used are of spectral grade purity with low viscosity. The effect of scattered light and background for each solvent was checked by using pure solvent in the cuvette and setting the excitation and emission monochromators at values used in polarization measurements. The contribution of this effect to the intensity of luminiscence was found to be less than 0·5%. The results obtained for polarization are given in Table I. The data on the excitation wavelength (λ_{ex}), the emission wavelength (λ_{em}) and the relative intensity of luminiscence in different solvents are also included in Table I.

The observed variation of polarization (Table I) cannot be accounted for as viscosity effect because in this case a 'Perrin Plot'[1] will not be linear. Also as the concentration is low the electromagnetic coupling of neighbouring molecules and its effect on decrease in polarization due to energy migration is small. In any case the concentration is the same in all the solvents and hence the variation in polarization cannot be explained only by energy migration. It is, therefore, likely that the observed variation in polarization is due to the dipole moment/dielectric constant of the solvent. The effect of these can be explained as follows: During absorption of the exciting radiation which takes place in a very short time, the equilibrium orientation of the dipole moment of the solute molecule, under the influence of dipole-dipole interaction, in the solvent cage of the surrounding solvent molecules, remains unchanged. But the solute molecules get enough time before a radiative transition takes place to reorient itself in the cage to get a new equilibrium state and this causes fluorescence depolarization. The Brownian rotation tends to destroy this equilibrium orientation and causes further depolarization. But the Brownian rotation is opposed by the dipole-dipole interaction between solvent-solute molecules which for a given solute depends on the dipole moment and the dielectric constant of the solvent. Thus this interaction tries to reduce the depolarization caused by the Brownian rotation. Therefore the polarization is expected to be higher in a solvent with higher dielectric constant/dipole moment. For acetone the dipole moment is comparable to water but the percentage polarization is lower. However as can be seen from Table I the intensity of fluorescence in this case is lower indicating some migration of energy which may cause depolarization. This additional cause of depolarization also explains the difference between the values of polarization for methanol and ethanol. Also the polarization should be minimum in non-polar solvents. To check this in the present case several non-polar solvents, *viz.*, benzene, cyclohexane, carbon tetrachloride; carbon disulphide, etc., were tried, but fluorescein is found

TABLE I

Solvent	Dipole[4] moment	Dielectric[4] constant	Viscosity[4]	λ_{ex} nm	λ_{em} nm	Relative intensity	% Polarization
Water	2·0–3·0	81	1·002	506	520	80	4·4
Methanol	1·65	32	0·594	512	524	81	2·0
Acetone	3·0	21	0·324	528	540	36	1·9
Ethanol	1·7	26	1·197	514	528	62	1·2

to be insoluble in these solvents. The results given in Table I clearly indicate the effect of dipole mement/dielectric constant on polarization of luminiscence.

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1. Perrin, F. J., *Phys. Radium*, 1926, 7, 390.
2. Weber, G., *Advances in Protein Chemistry*, 1953, 8, 415; 1960, 75, 335.
3. Rathi, S. S. and Machwe, M. K., *Physics Letters*, 1967, 25 (1) A, 41.
4. Kaye, G. W. C. and Laby, T. H., *Tables of Physical and Chemical Constants* (Longmans and Green Co., Ltd., London), 1966.

STUDIES ON THE REACTION OF ZIRCONIUM (IV) WITH 4-METHYL-DAPHNETIN

Only a few studies have been carried out on Zirconium (IV) complexes[1] with O-diphenols and no work on Zirconium complexes with O-dihydroxy coumarins has been reported.

Zirconium (IV) reacts with 4-methyl daphnetin (4-methyl, 7-8-dihydroxy coumarin) giving a pale yellow colour. In the present work this colour reaction of Zr(IV) with 4-methyl daphnetin has been studied and the nature of the different species which exist in solution at different pH values and metal ion and ligand concentration has been elucidated.

Experimental: Stock solution of Zirconium (IV) was prepared by dissolving $ZrOCl_2.8H_2O$ (E. Merck pro. analyst) in HCl and diluted with distilled water. 4-Methyl-daphnetin was prepared by the method of Pechmann[2] and recrystallised twice from dilute ethanol. All other chemicals used were of analytical grade. A Beckmann pH meter type zeromatic and a colemann spectrophotometer (model 14) were used. All the investigations have been carried out in 50% aqueous-ethanol medium. The absorbance of the ligand in excess was always substracted from all readings neglecting the part of the ligand complexed.

Results:

It was found that the reaction of Zr (IV) with 4-methyl-daphnetin is reversible in nature and the absorbance of the complex remains unchanged for over 2 days.

The pH-absorbance curves of Zr (IV)-4-methyl-daphnetin complex at various metal ion concentration showed that only mononuclear complexes were formed in solution[3]. Further investigations were carried out to find out the composition and nature of different complexes formed in solution. A family of curves was obtained with variations in ligand concentration at different pH values maintaining a fixed concentration of the metal ion. These pH absorbance curves have been interpreted by logarithmic method of analysis[3] and the method of corresponding solutions[3]. The method of logarithmic analysis showed the existence of 1 : 1 complex in the pH region 1·0–2·8 and a 1 : 2 complex in the pH region 3·4–3·8. It also showed that only one molecule of 4-methyl-daphnetin is successively coordinated and only one proton is liberated in each step of complexation in solution with large ligand excess. The same has been confirmed by the method of corresponding solutions.

The following equilibrit can be assumed in the above pH region:

$$ZrO^{+2} + H_2R \rightleftharpoons ZrO(RH)^+ + H^+$$
$$ZrO(RH)^+ + H_2R \rightleftharpoons ZrO(RH)_2 + H^+.$$

The composition of the two complexes which are formed in different pH regions was also established by the method of continuous variations[4]. After pH 4·0 precipitation started, and further study was discontinued.

The following values for stability constants and molar absorptivities of the two complexes have been obtained.

Table I

Constants	Equilibrium	log K	Molar absorptivities $\epsilon \times 10^{-3}$ (360 nm)
k_1	$[M(RH)][H]/[M][H_2R]$	1·8290	4·95
k_2	$[M(RH)_2][H]/[M(RH)][H_2R]$	−0·8378	5·67

Authors express their grateful thanks to Dr. Yag Dutt, for his valuable suggestions and constant encouragement.

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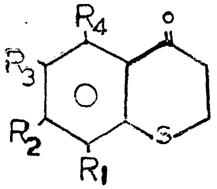
1. Sommer, L. and Bartusek, M., *Folia., Fac., Nat. Univ. Brno.* (*Chemia*), (VII), 1966.
2. Pechmann, H. V., *Ber.* 1884, 17, 929.
3. Sommer, L., Kuban, V. and Havel, J., *Folia., Fac. Rer. Nat. Univ. Brno.* (*Chemia*), (XI), 1970.
4. Job, P., *Ann. Chim.*, 1928, 9, 113.

SYNTHESIS OF SOME THIOCHROMANONES

In continuation of our previous paper on the synthesis of α-pyronochromanones, the present work describes the S-cyanoethylation of the different thiocresols leading to the synthesis of thiochromanones.



I, $R_1 = CH_3$; $R_2 = R_3 = 4_4 = H$.
II, $R_4 = CH_3$; $R_1 = R_2 = R_3 = H$.
III, $R_2 = CH_3$; $R_1 = R_3 = R_4 = H$.
IV, $R_3 = CH_3$; $R_1 = R_2 = R_4 = H$.

The reaction of *o*-thiocresol with acrylonitrile in the presence of aq. NaOH (10%) in boiling dioxane for 15 hrs. yielded the corresponding mercaptopropionitrile[1] as a light yellow oil (b.p. 177°/18 mm) which was hydrolysed by heating with conc. HCl (1 hour) to the propionic acid, crystallised from benzene-petroleum ether in colourless needles, m.p. 93–95° (Found: C, 60·8; H, 6·1%; $C_{10}H_{12}O_2S$ requires: C, 61·2; H, 6·1%). Cyclisation of the above acid on heating with conc. H_2SO_4 on a water bath for about 10 minutes afforded 8-methyl-thiochromanone (I) crystallised from benzene-petroleum ether in colourless needles, m.p. 65–70° (Found: C, 67·3; H, 5·9%. $C_{10}H_{10}OS$ requires: C, 67·4; H, 5·6%). The NMR (CCl_4) spectrum was fully consistent with its structure and showed signals at δ 2·3 (3 H, s, $-CH_3$ at C_8) δ 3·5 (4 H, m, $-CH_2$ at C_2 and CH_2 at C_3); δ 7·1 (2 H, m, at C_6 and C_7); δ 7·95 (1 H, d, at C_5, J = 2 Hz and 6 Hz). The 2, 4-DNP from (I) had m.p. 238–39° (Found: N, 15·5%: $C_{16}H_{14}O_4N_4S$ requires: N, 15·6%).

The mercaptonitrile from *m*-thiocresol by cyanoethylation under the above conditions was a yellow oil (b.p. 175–78°/18 mm). and gave the mercapto acid as colourless needles from benzene-petroleum ether, m.p. 68–70° (Found: C, 61·1; H, 6·4%. $C_{10}H_{12}O_2S$ requires: C, 61·2; H, 6·1%). Cyclisation of the latter acid with conc. H_2SO_4 yielded a reddish brown oil which was chromatographed over neutral alumina. The fraction eluted with petrol ether was a yellow oil (b.p. 124°/2·5 mm) which was assigned the structure as 5-methyl-thiochromanone (II) on the basis of its NMR data (Found: C, 67·3; H, 5·6%. $C_{10}H_{10}OS$ requires: C, 67·4; H, 5·6%). NMR (CCl_4); δ 2·55 (3 H, s, $-CH_3$); δ 3·0 (4 H, m, $-CH_2$ at C_2 and $-CH_2$ at C_3) at δ 6·95 (3 H, m, at C_6, C_7 and C_8) 2, 4-DNP of II, m.p. 206° (Found: N, 15·4%. $C_{16}H_{14}O_4SN_4$ requires: N, 15·6%). The fraction eluted with petrol ether-benzene (1 : 4) was a reddish oil, b.p. 146°/2·5 mm (Found: C, 67·1; H, 5·4%. $C_{10}H_{10}OS$ requires: C, 67·4; H, 5·6%). Its 2, 4-DNP had m.p. 223° (Found: N, 15·7%. $C_{16}H_{14}O_4SN_4$ requires: N, 15·6%). Its structure as 7-methyl-thiochromanone (III) was in complete agreement with its NMR spectrum (CCl_4): δ 2·35 (3 H, s, $-CH_3$). δ 3·0 (4 H, m, $-CH_2$ at C_2 and $-CH_2$ at C_3); δ 6·90 (1 H, d, at C_8, J = 2 Hz); δ 6·98 (1 H, dd, at C_6, J = 8 Hz and 2 Hz); δ 7·9 (1 H, d, at C_5, J = 8 Hz).

By the cyanoethylation of *p*-thiocresol, the required propionitrile was obtained as a yellow oil[1] (b.p. 174°/18 mm) which was easily hydrolysed with HCl to give the corresponding acid as colourless needles, m.p. 70° from benzene-petrol ether. On cyclisation, the acid afforded 6-methyl-thiochromanone (IV) as colourless needles, m.p. 41°[2], 2, 4-DNP, m.p. 285–86° (Found: N, 15·8%. $C_{16}H_{14}O_4N_4S$ requires: N, 15·6%). NMR($CDCl_3$) of IV: δ 2·3 (3 H, s, $-CH_3$); δ 2·9–3·1 (4 H, m, methylene protons at C_2 and C_3); δ 7·15 (2H, s, at C_7 and C_8). δ 7·9 (1 H, s, at C_5).

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Institute of Science, Mrs. S. M. Thakkar.
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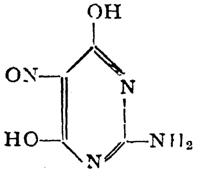
Bombay-32, *August* 8, 1975.

---

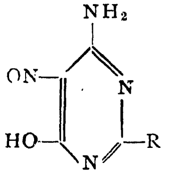
1. Hurd, C. D. and Gerahbein, L. L., *J. Am. Chem. Soc.*, 1947, 69, 2328.
2. Arndt, F., Flemming, W., Scholz, E. and Lowenson, V., *Ber.*, 1923, 56B, 1279.

IRON(II) AND COBALT(II) COMPLEXFS WITH PYRIMIDINES

In a recent communication[1], analytical characteristics of 2-amino-5-nitroso-4, 6-pyrimidinediol (I) have been reported. It was thought of interest to examine the analytical potentialities of related pyrimidines (II and III) and to study the effect



(I)



(II) R=CH_3; ('II) R=SH

TABLE I

Characteristics of metal complexes with pyrimidines

Characteristic	Fe^{2+}-complex with		Co^{2+}-complex with	
	(II)	(III)	(II)	(III)
Colour	Bluish-green		Yellow	
λ_{max} (nm)	675	650	370	420
ϵ_{max} ($l \cdot mole^{-1} \cdot cm^{-1}$)	12,000	14,600	19,440	48,000
Pyrimidine needed for full colour development	10-Molar concent.	53-Molar concent.	30-Molar concent.	40-Molar concent.
pH range for maximum and const. absorbance	5·0–8·8	6·7–9·3	3·0–5·5	6·5–8·8
Beer's law range (ppm)	Up to 5·4	Up to 3·3	Up to 2.4	Up to 1·3
Sandell's sensitivity	0·0047	0·0035	0·0054	0·0012
Composition (metal : reagent)	1 : 3	1 : 2	1 : 2	1 : 3
Behaviour in organic solvents	Not extractable		Not extractable	
Standard deviation (with 6 samples)	0·003	0·001	0·001	0·002

TABLE II

Toleration limits (ppm) of foreign ions

Foreign ion	Iron determination with		Cobalt determination with	
	(II)	(III)	(II)	(III)
Fluoride	200	I	500	1000
Chloride	1000	1000	1000	1000
Bromide	1000	500	1000	1000
Iodide	1000	I	400	1000
Nitrite	200	I	1000	I
Perchlorate	1000	300	1000	1000
Thiocyanate	1000	500	1000	1000
Acetate	1000	I	1000	1000
Thiosulphate	500	I	500	1000
Oxalate	30	I	I	1000
Tartrate	100	500	I	1000
Thiourea	1000	I	1000	100
Borate	1000	I	500	1000
Phosphate	I	I	1000	1000
Citrate	50	I	1000	1000
Calcium (II)	100	100	100	100
Strontium (II)	100	100	100	100
Barium (II)	150	60	150	150
Zinc (II)	350	I	70	I
Cadmium (II)	300	I	60	I
Mercury (II)	I	I	50	I
Indium (III)	20	100	50	I
Thallium (I)	100	I	I	200
Tin (II)	50	I	I	100
Antimony (III)	150	I	120	120
Vanadium (IV)	5	5	5	5
Molybdenum (VI)	I	70	20	50
Manganese (II)	50	I	50	I
Ruthenium (III)	I	I	I	2

I stands for interfere.

The presence of copper (II), cerium (IV), nickel (II), osmium (VIII) and palladium (II) ions causes interference in determinations.

of other substituents in the molecule. Standard solutions of 0·005 molar strength of II (Aldrich Chemical Co., U.S.A.) and III (K and K Laboratories, U.S.A.) were prepared by dissolving the appropriate amounts in hot water and hot HCl (1 N), respectively. Other solutions were prepared as reported earlier[1]. The pH of the solutions were adjusted with acetate buffers and dilute solutions of NaOH and HCl.

Light yellow coloured solution of (III) has absorption maxima at 260 ($\epsilon = 7480$) and 330 nm ($\epsilon = 6720$) whereas for (II) the absorption maxima lie at 300 ($\epsilon = 4790$) and 345 nm ($\epsilon = 6420$). The characteristics of iron(II) and cobalt(II) complexes formed with the two pyrimidines are summarized in Table I.

Determination of Iron (II) or Cobalt (II)

To a suitable aliquot of iron (II) or cobalt (II) solution, sufficient excess of pyrimidine (II) or (III) was added, pH adjusted in accordance with Table I and the solution finally diluted ta a fixed volume. The absorbance was measured at λ_{max} of the species against appropriate reagent blank. The amount of the metal was calculated by comparison of the absorbance with a pre-calibrated graph.

The toleration limits (in ppm) of diverse ions were found in determination of 2·8 ppm of iron and 1·4 ppm of cobalt with pyrimidine (II) and 1·4 ppm of iron and 0·73 ppm of cobalt with pyrimidine (III). The results are incorporated in Table II.

From the results (Table I), it may be concluded that the sensitivity of the thiol substituted pyrimidine (III) is more and it forms metal complexes at comparatively high pH range. On the other hand, methyl substituted pyrimidine (II) acts more selectively in determination of iron and many ions do not interfere (Table II).

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1. Kundra, S. K., Katyal, M. and Singh, R. P., *Curr. Sci.*, 1975, **44**, **81**.

EFFECT OF VARYING WATER REGIMES ON STEM ELONGATION OF DEEP WATER RICE VARIETIES

An investigation was carried out on stem elongation of two photosensitive deep water rices, *viz.*, *Jaladhi* 1 and *Jaladhi* 2, under four depths of submergence at Rice Research Station, Chinsurah, West Bengal.

Water regimes	Maximum water level (cm)	
	Vegetative phase	Reproductive phase
(*a*) Shallow water	20	15
(*b*) Knee-deep water	50	35
(*c*) Semi-deep water	115	85
(*d*) Deep water	180	140

The seeds were directly sown by the end of April and harvested from end of November to December. The average data of 5 replications on the length of internodes of main shoot at harvest were graphically represented in Fig. 1.

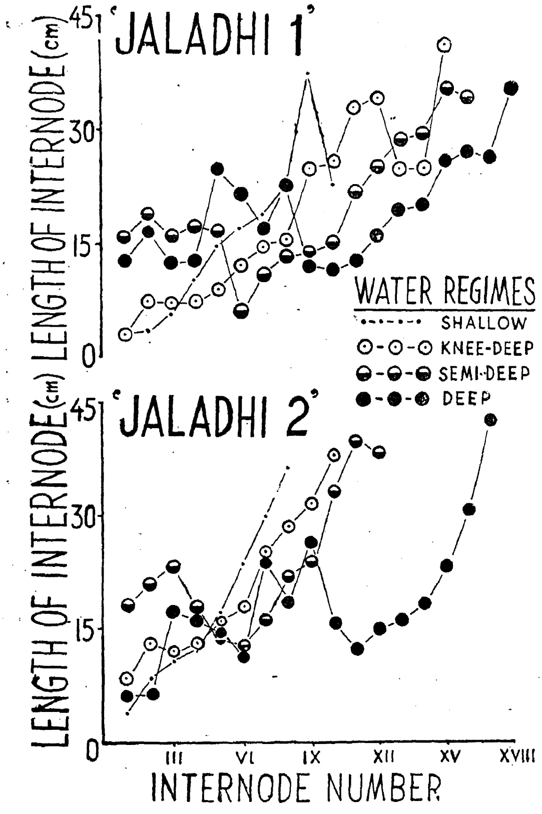

Fig. 1. Length of internode (base to apex) of two deep water rice varieties under four water regimes.

The total stem length increased remarkably with the rise of water level, owing to the formation of appreciably larger number of longer internodes in both the varieties. The length of different internodes varied considerably, owing to the occurrence of variable depths of water present during the vegetative as well as reproductive phases of plant growth. A similar result was noticed by Majid[1], in the deep water varieties, where a quicker rise of water level resulted in longer internodes. The results also indicated that important characteristic of the water-loving varieties was the capacity of the plants to elongate their culms with the rise of water level, so that the top leaves remained near or above the surface of the water. In general, the internode elongation of the varieties varied with the degree of submergence. As regards growth in stem length and number of internodes under shallow water condition, the plants showed remarkably reduced values and behaved similar to normal tall *indica* rice varieties (Fig. 1). But appreciable higher values of total stem length obtained from the knee-deep water and the semi-deep water conditions which were much high in comparison to their respective depth of watering treatments, clearly points out that due to their floating behaviour the stems have a tendency to grow along the surface of water.

There was a slight difference in the elongation between the two varieties under different depths. Irrespective of treatments, *Jaladhi* 1 exhibited better performance in relation to stem elongation, as it showed maximum extension growth of culm under deep water condition attaining 342 cm stem length per plant at harvest which was 10·1% greater than *Jaladhi* 2. Moreover, the maximum number of internodes in the main shoot was more (18) than in *Jaladhi* 2 (17) under deep water conditions. Data suggest the total stem length that a particular variety is capable of attaining is one of the important criteria which could be used for selecting varieties for different depths of water condition.

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Salt and Flood Resistant Paddy Scheme,
Rice Research Station,
Chinsurah, West Bengal, *May* 9, 1975.

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1. Majid, S., *Annual Report of the Deep Water Research Station, Habiganj, Assam*, 1936–37.

A DENSITY GRADIENT METHOD FOR FRACTIONATION OF BUFFALO SPERMATOZOA

In an earlier report[1], fractionation of buffalo spermatozoa was achieved using the basic principles of homogenization and fractionation with different molar concentrations of sucrose. However, the procedure was inconvenient and amenable to morphological alterations due to longer time of homogenization. In the present communication, a modified and simple procedure is described which is based on the principles of ultrasonication and density gradient centrifugation.

Approximately 2 ml of freshly collected buffalo semen was centrifuged at 1400 × g for 10 minutes. Seminal plasma was decanted. The sperm pack was resuspended in an equal volume of Ca^{++} -free Ringer solution, and centrifuged again for 10 minutes at 1400 × g. The process was repeated twice. The washed sperm pack was diluted three fold with distilled water and disintegrated with the aid of Vibronics ultrasonicator (model VPL-P1) for 60 seconds at a frequency of 25 kilocycles/second.

A density gradient of sucrose was prepared by layering 1 ml each of 50%, 40%, 30% and 20% sucrose in the ascending order in the centrifuge tube (29 mm × 116 mm). An amount of 0·7 to 1 ml of ultrasonicated sperm was layered on the top of sucrose gradient and centrifuged at 1050 × g for 8 minutes. After centrifugation, two distinct layers were obtained and sizable amount settled at the bottom of centrifuge tube. The two layers were carefully removed with the aid of pasteur pipette.

Smears of the two layers and sediment were stained with Rose Bengal and examined microscopically. The first layer consisted of tails (tail fraction) which were fragmented almost to collodial dimensions and hence its morphological entity could not be maintained. The second layer comprised of mid pieces (mid piece fraction) and the sediment consisted of sperm heads (head fraction). In order to ascertain the purity of different fractions, smears were prepared from each fractions and cytochemically assayed for the succinic dehydrogenase activity by the method of Hrudka[2]. Microscopical examination revealed that in the ultrasonicated material and the different fractions, only the mid piece showed an intense activity of succinic dehydrogenase which was in agreement with the findings of Hrudka[2]. The purity of different fractions was more than 95%. In some samples, however, it was observed that the contamination of mid pieces in head fraction was more than 10%. In such cases, the head fraction was resonicated for 15 seconds and the same procedure was followed in order to achieve purity of more than 95%.

An attempt was made to study the recovery of protien from standard preparation (S–P), *i.e.*, ultrasonicated sperm, in the different fractions. The protein was estimated by method of Lowry *et al.*[3]. Table I shows the relative distribution of protein in different fractions as compared to S–P. The recovery was 95·1%.

TABLE I

Recovery of proteins in different fractions

	Mg of protein	Relative %
S-P	10·010	100 00
Head	1·732	17·30
Mid piece	0·998	9·97
Tail	6·790	67·83
Recovery	9·520	95·1

The optical density of the S–P and different fraction was determined at 260 mμ and 280 mμ. Table II gives the ratio 280/260. A relatively lower value of 280/260 ratio for head fraction further confirms the purity of the fraction since the head comprised mainly of nucleic acids.

TABLE II

Ultraviolet absorption of different fractions

	$O.D._{260 m\mu}$	$O.D._{280 m\mu}$	280/260 ratio
S-P	0·585	0·244	0·417
Head	0·668	0·194	0·290
Mid piece	0·721	0·620	0·859
Tail	0·658	0·585	0·88

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Jabalpur (M.P.), India, *July* 12, 1975.

1. Chauhan, R. A. S. and Srivastava, R. K., *Ind. J. Exptl. Biol.*, 1974, 12, 501.
2. Hrudka, F., *J. Reprod. Fertil.*, 1965, 10, 15.
3. Lowry, O. H., Rosebrough, N. J., Farr, A. L. and Randall, R. J., *J. Biol. Chem.*, 1951, 193, 265.

CYCLIC NUCLEOTIDE PHOSPHODIESTERASE OF *HARTMANNELLA CULBERTSONI*

Previous studies from this laboratory have implicated cyclic 3′ : 5′ adenosine monophosphate (cyclic AMP) through its intracellular concentration to regulate the biochemical events leading to the differentiation of trophic *Hartmannella culbertsoni* into viable cysts[1-3]. The intracellular concentration of cyclic AMP at any phase of cellular life reflects the turnover of this regulatory nucleotide involving adenylate cyclase mediating its synthesis and cyclic nucleotide phosphodiesterase catalysing its degradation[4]. Biogenic amines which trigger encystation of amoeba also activate its adenyl cyclase[5]. Some properties of the cyclic nucleotide phosphodiesterase are described in this communication.

Hartmannella culbertsoni[6], obtained from the collection of Dr. B. N. Singh of this Institute was grown in 1% (w/v) proteose peptone, 1% (w/v) tryptone and 0·5% (w/v) NaCl, pH 6·8 as described elsewhere[7]. Six-day growth of cells was harvested by centrifugation at 800 g and washed by dispersal in sterile 150 mM NaCl and centrifugation at 800 g. Homogenates (10% w/v) were prepared in cold 250 mM sucrose by grinding the cells for 1–2 min in a Potter-Elvehjem glass homogenizer fitted with a Teflon pestle and setting the motor speed at 4,000 rpm. The homogenates were subjected to differential centrifugation.

cAMP phosphodiesterase was assayed according to the procedure described by Butcher and Sutherland[8] and inorganic phosphorus estimated in the supernatant by the procedure described by Fiske and Subba Row[9].

By differential centrifugation about 60% of the enzyme activity was recovered in the 100,000 g supernatant (organelle free cytosol) and the rest of the activity was distributed almost equally among fractions sedimenting at 800 g, 15,000 g and 100,000 g.

The crude enzyme was active on the free acid form of cyclic AMP as well as on its dibutyryl derivative. On preincubation, theophylline exerted 40% inhibition at 1 mM and 80% inhibition at 10 mM. The enzyme was stimulated 40% by 40 mM imidazole.

The activity of the enzyme was followed in cells as well as the medium used for starving and triggering differentiation. Some typical results are summarised in Table I. The activity of cells exposed to taurine or epinephrine during starvation was significantly less than of the control cells in plain starvation medium. Some amount of enzyme was also secreted into the medium irrespective of whether the cells were in plain starvation medium or in the taurine or epinephrine supplemented medium.

The decay of cAMP phosphodiesterase under starvation conditions suggests that intracellular concentration of cyclic AMP could increase. The decay of the enzyme is faster when cells are committed to differentiation in the starvation medium by the triggering action of taurine or epinephrine. Earlier it has been shown that the activity of adenyl cyclase of *H. culbertsoni* remains unchanged during six hours of starvation whereas it is stimulated 2–3 fold in the same cells undergoing differentiation[1]. Thus the stimulated adenylate cyclase and the fast decaying cAMP phosphodiesterase together appear to be responsible for the build-up of a relatively high intracellular level of cAMP in the differentiating amoeba as compared to merely starving amoeba.

TABLE I

cAMP phosphodiesterase activity in cells and medium

Time of exposure (hr.)	Control medium		Encystment medium: Taurine medium		Encystment medium: Epinephrine medium	
	C	M	C	M	C	M
			Enzyme units*			
0	262	0	262	0	262	0
1	240	19	217	15	229	15
2	217	30	172	26	165	30
3	157	30	131	23	101	23
4	135	34	101	19	75	23
5	127	30	90	23	68	19
6	124	24	75	15	56	15

Assay mixtures in a final volume of 0·8 ml contained 0·36 μ mole cyclic AMP, 36 μ moles acetate buffer, pH 5·6, 1·8 μ moles $MgSO_4$ and enzyme. After 60 min. incubation at 37° C the contents were held at the temperature of boiling water for 2 min. and cooled. The pH of reaction mixture was adjusted to 7·5 with 0·1 N NaOH and 0·1 ml of a solution of cobra venom (Naja naja; from Haffkine Institute, Bombay) (1 mg/ml of Tris HCl buffer, pH 7·5) was added and incubated for 30 min, and the reaction stopped by the addition of 0·1 ml 55% w/v trichloroacetic acid, centrifuged and supernatant used for Pi estimation.

* Values are enzyme units in 1×10^7 cells (1 unit ≡ amount of enzyme that cleaves 1 *n* mole of cyclic AMP in 60 min).

C = cells; M = medium; Control cells in 86 mM NaCl. Encystment medium = cells in medium containing 15 mM taurine or 0·5 mM epinephrine, 20 mM $MgCl_2$, and 86 mM NaCl.

Aliquots of cells (2×10^7) were taken at indicated intervals, separated from medium and both cells and medium used in assay of enzyme activity.

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1. Raizada, M. K. and Krishna Murti, C. R., *J. Cell. Biol.*, 1972, 52, 743.
2. Krishna Murti, C. R., *Curr. Sci.*, 1971, 40, 589.
3. —, *Biochem. Soc. Trans.*, 1973, 1, 1104.
4. Robison, G. A., Butcher, R. W. and Sutherland, E. W., *Cyclic AMP*, Acad. Press, New York, 1971, p. 84.
5. Verma, A. K., Raizada, M. K. and Krishna Murti, C. R., *Biochem. Pharmacol.*, 1974, 23, 57.
6. Singh, B. N. and Das, S. R., *Phil. Trans. Roy. Soc.* (Lond.), 1970, 259 B, 435.
7. Raizada, M. K. and Krishna Murti, C. R., *J. Protozool.*, 1971, 18, 115.
8. Butcher, R. W. and Sutherland, E. W., *J. Biol. Chem.*, 1962, 237, 1244.
9. Fiske, C. H. and Subba Row, Y., *Ibid.*, 1925, 66, 375.

FOSSIL VERTEBRATES FROM MARUVATTOOR (TIRUCHCHIRAPPALLI DISTRICT, TAMIL NADU)

II. Notes on the Equine and Bovine Teeth

THE present note deals with two fossil teeth belonging to the genera *Equus* Linnaeus and *Bos* Linnaeus collected by Dr. K. V. Lakshminarayana, in 1971 from the alluvial beds on the bank of Marudiyar River near Maruvattoor Tiruchchirappalli District, Tamil Nadu.

These mammalian remains might have been drifted by the nullahs flowing over some adjoining Late Pleistocene deposits on the Cretaceous rocks (*vide* Mamgain and Sastry, 1967). A brief description of the fossil teeth are given below.

Equus sp.
(Plate 1, Fig. A)

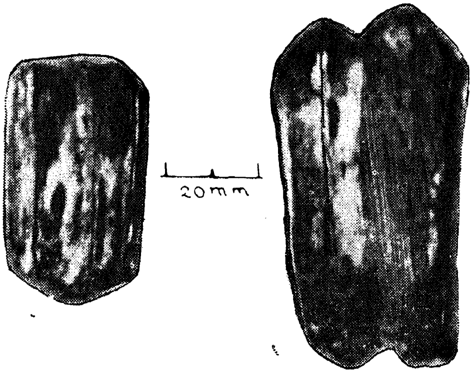

PLATE I. Fig. A. *Equus* sp. [Lower right 3rd premolar]. Fig. B. *Bos* sp. [Upper left 2nd molar].

Material

Lower right 3rd premolar (Reg. No. V.P. 3).

Description

Angle between metaconid and metastylid V-shaped, protoconid and hypoconid rather straight; parastylid not reaching metaconid; length of the crown 25·5 mm; total height 42 mm; maximum breadth of the crown 14·5 mm.

Remarks

The structure of the tooth resembles largely that of *Equus namadicus* Falc. and Caut., but nothing definite can be said about its specific status due to the absence of upper tooth and other supporting data. However, Rao and Seshachar (1927) and Rao (1927) have also reported fossil equine tooth from Ariyalur, Tamil Nadu.

Bos. sp.
(Plate 1, Fig. B)

Material

Upper left second molar (Reg. No. V.P. 4).

Description

Hypsodont; erupting tooth having basal pillar, tip of the basal pillar 12 mm below the tip of the crown; length of crown 33 mm; height of tooth 64 mm; maximum breadth of the crown 17·2 mm.

Remarks

Mamgain and Sastry (1967) reported the occurrence of *Bos* sp. from Ariyalur, based on lower jaw and limb bones. The present is, therefore, the first record of an upper tooth, hence interesting.

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Zoological Survey of India, K. D. SAHA.
8, Lindsay Street,
Calcutta 700 016, *September* 23, 1974.

1. Mamgain, V. D. and Sastry, M. V. A., *Indian Miner.*, 1967, 21 (1), 61.
2. Rao, C. R. N. and Seshachar, B. R., *Half-yearly J. Mysore Univ.*, Bangalore, 1927, 1 (2), 144.
3. Rao, L. R., *Ibid.*, 1972, 1 (2); 153.

A NOTE ON THE SMALLER FORAMINIFERA FROM THE BARIPADA BEDS, MAYURBHANJ DISTRICT, ORISSA

THE present note deals with the foraminifera obtained from the Baripada Beds exposed on the eastern bank of the Burhabalang river. The writers collected the samples from the Baripada Beds during 1970 and observed the following stratigraphic sequence of the rocks :

	Lithounits	Age	Thickness in metres
	Laterite Conglomerate	Post-Miocene	4· 1·
Baripada Beds	Greenish grey shales	Middle Early Miocene —? Late Miocene	6
	Arenaceous yellowish white fossiliferous limestone containing megafossils—*Ostrea* sp., etc.	Early Mioeene	1·2
	Greenish white shales	Early Miocene	Not determined

The greenish grey shales have yielded a rich assemblage of species of many smaller foraminifera. The most important among them are: *Ammonia beccari Kaeboeensis* (Le Roy), *Cribroelphidium subincertum* (Asano), *Turborotalia obesa* (Bolli), *Turborotalia continuosa* (Blow), *Florilius communis* (d'Orbigny), *Buliminella brevior* Cushman, *Buliminella hanzawai* Asano, *Buliminella* aff. *B. longicamerata* Bandy, *Brizalina singhi* n. sp., *Valvulineria sastrii* n. sp., *Cribrononion dattai* n. sp., *Lagena amphora,* and *Cibicides tewari* n. sp. Other forms identified upto generic level are *Triloculina, Uvigerina, Lagena* etc. *Ammonia beccari Koeboeensis* (Le Roy) occurs in abundance in the arenaceous yellowish white fossiliferous limestone.

Blow[1] recorded the range of *Turborotalia obesa* from within the Zone N_2 to Zone N_{23} (Late Oligocene to Pleisto-Holocene). It is, therefore, a long ranging form. *Turborotalia continuousa,* on the other hand, is restricted within the Miocene. Blow[1] considers its range from within Zone N_6 to Zone N_{17} (middle part of Early Miocene to Late Miocene). In view of this, the greenish grey shales may be referred to the middle Early Miocene to Late Miocene, with the conformably underlying limestone and greenish white shales lying very likely at the base of the Early Miocene.

The valuable suggestions of Prof. S. N. Singh and Prof. Don L. Eicher, Department of Geological Sciences, Colorado University, are gratefully acknowledged. One of the writers (A. K. J.) is thankful to the Ministry of Education, Government of Uttar Pradesh, for financial assistance in the year 1970.

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1. Blow, W. H., *Proc. First Int. Conf. Plankt. Micro.,* 1967, 1, 199.

THE MORPHOLOGY OF *MONOCERCOMONOIDES CHAKRAVARTII* N. SP. (PROTOZOA: MASTIGOPHORA) FROM THE GUT OF THE INSECT *POLYPHAGA INDICA*

DURING the course of a survey of the intestinal flagellates of arthropods of Maharashtra, an interesting flagellate was found in the middle and hind gut of the insect *Polyphaga indica.* The organism, belonging to the genus *Monocercomonoides* Travis (1932)[1], was relatively rare in its occurrence and was found in only a few of the 249 hosts examined.

In the stained preparations, the parasite appears somewhat spherical (Figs. 2, 7, 8); pyriform (Fig. 1) or irregular (Figs. 3, 5) in outline. The blepharoplastic complex situated at the anterior end of the body comprises of three granules (Figs. 4, 7, 8). One of these gives origin to the funis, while the second one is the point of origin of two anterior flagella and the third one gives rise to one anterior flagellum, the trailing flagellum and the axostyle (Figs. 4, 7, 8).

The three anterior flagella are unequal, measuring 4·6–11·8 μm, 6·7–12·8 μm and 7·2–14·9 μm with averages of 7·4 μm, 8·8 μm and 9·8 μm respectively. The trailing flagellum is about twice the length of the body and measures 9·8–21·6 μm (average 14·5 μm). In many cases one of the flagella is thicker than the others (Figs. 3, 4, 6). Some of the flagella terminate in distinct acronemes (Figs. 5, 8). The funis is short and filamentous (Figs. 4, 7, 8) and measures 2·6–4·6 μm, with the average of 3·6 μm.

The axostyle is extremely delicate and could be observed only in a few well stained organisms (Figs. 1, 2, 5). It extends upto the posterior end of the body (Fig. 1) or projects out slightly (Figs. 2, 5). The posterior half of the axostyle is slightly thicker than the anterior half (Fig. 5). The posterior tip is enlarged into a fine spear-shaped structure, tapering to a point (Figs. 2, 5). The nucleus lies behind the blepharoplastic complex and is variable in shape and size. It is

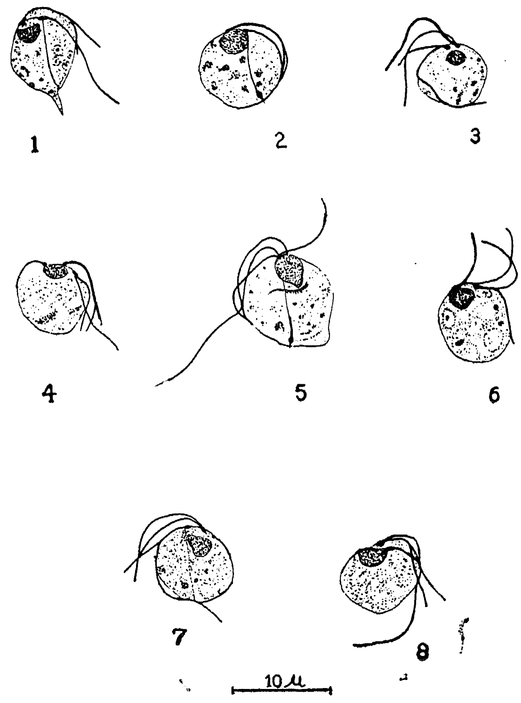
TABLE I

Showing a comparison of the various species of the genus Monocercomonoides *from insects*

S. No.	Species	Body dimensions (in μm)	Blepharo-plasts	Funis	Axostyle
1.	*Monocercomonoides melyolonthae* Grassi, 1879	6·2–12·5 × 4·3–10·0 (10·0×7·5)	2	Absent	Slender, upto posterior end
2.	*M. orthopterorum* Parisi, 1910	3·7– 7·0 × 2·1– 3·8 (6·2×3·1)	2	Absent	Very fine, sometimes not recognisable
3.	*M. garnhami* Bhaskar Rao, 1962	5·0–12·5 × 4·5–14·5 (9·1×8·3)	2	Long (7·5 μm)	Thin proximally, thick distally; limited to body wall
4.	*M. quadrii* Bhaskar Rao, 1969	4·5–16·5 × 4·5–17·5 (11·1×10·9)	2	Absent	Thick in middle, fine posteeriorly
5.	*M. ganapatii* Bhaskar Rao, 1969	6·0– 9·0 × 4·5– 8·5 (7·4×6·6)	2	Short filamentous (3·08 μm)	Longer than body; uniform in thickness; posterior tip knob-like
6.	*M. krishnamurthii* Sultana (In press)	5·7– 9·8 × 3·6– 7·2 (7·4×5·2)	2	Absent	Filamentous; surrounded by typical peri-axostylar vacuole near posterior end
7.	*M. chakravartii* n. sp.	5·7– 9·8 × 5·7–13·4 (7·4×7·2)	3	Short filamentous (3·6 μm)	Delicate; posterior tip typically spear-like

broader than long and measures 2·1 × 2·6 μm on the average, though it appears spherical in a few individuals (Fig. 2). The cytoplasm stains homogeneously and has deeply staining granules and bacteria. The periplast is thin. The dimensions of the body of the organism are 5·7–9·8 × 5·7–13·4 μm (average 7·4 × 7·2 μm).

Six species of this genus are so far described from insect hosts. These are *M. melolonthae* Grassi (1879)[2]; *M. orthopterorum* Parisi (1910)[3]; *M. garnhami* Bhaskar Rao (1969)[4]; *M. qadrii* Bhaskar Rao (1969)[4]; *M. ganapatii* Bhaskar Rao (1969)[4] (also redescribed by Sultana—In press)[5] and *M. krishnamurthii* Sultana (In press)[6]. The species described above shows distinct differences from all these species, as shown in Table I. Particularly characteristic of this organism are the presence of three granules in the blepharoplastic complex, the presence of a spear-like tip of the axostyle and one flagellum thicker than the rest. In view of these, it is considered new to science and designated *Monocercomonoides chakravartii* n. p., after Dr. M. M. Chakravarti, Retired Professor of Zoology, Calcutta University, Culcutta, India.



FIGS. 1–8. *Monocercomonoides chakravartii* n. sp. Fig. 1. Showing axostyle upto posterior end. Fig. 2. Showing spear-like axostylar tip. Figs. 3, 4, 6. Sowing one flagellum thicker than others. Fig. 5. Showing acronematic flagella and axostylar structure. Fig. 7. Showing blepharoplasts, origin of flagella and funis. Fig. 8. Showing general structure.

(All figures from smears exposed to osmic vapours, fixed in methanol and stained with Giemsa's stain—drawn at 2,000 ×).

The authors are grateful to Dr. R. Nagabhushanam, for providing laboratory and library facilities.

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Marathwada University, TAYYABA SULTANA.
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---

1. Travis, B. V., *Iowa State Coll. J. Sci.*, 1932, 5, 223.
2. Grassi, B., *Gaz. Med. Ital. Lombardi.*, 1879. 39, 445.
3. Parisi, B., *Arch. Protistenk.*, 1910, 19, 232.
4. Bhaskar Rac, T., *Proc. Ind. Acad. Sci.*, 1969, 70, 208.
5. Sultana, T., *Marathwada Univ. J. Sci.*, (In Press).
6. —, *Ibid.*, (In Press).

ON THE OCCURRENCE OF *BOS NOMADICUS* FALCONER, FROM THE PREHISTORIC SITE OF SUSUNIA, DISTRICT BANKURA, WEST BENGAL

THE Geological Survey of India in collaboration with the Archaeology Department of the Government of West Bengal, undertook a programme (1969) of exploration and excavation of prehistoric sites in the State, as a result of which Susunia and Beharinath in the North Western Bankura District yielded interesting prehistoric animal remains. These include a mandible of a bovine mammal identified as *Bos namadicus,* which forms the subject of the present paper.

The specimen (Fig. 1) is almost completely fossilized. The mandible is much broader than that of any living bovines. It is 40 mm. thick. The teeth are all hypsodont. The external lobes of the molar are prominent and the ribs of the posterior lobes are almost as strong as those of the anterior lobes. The crescentic Islands are not wide. The teeth possess comparatively less cement than in species of *Bubalus.*

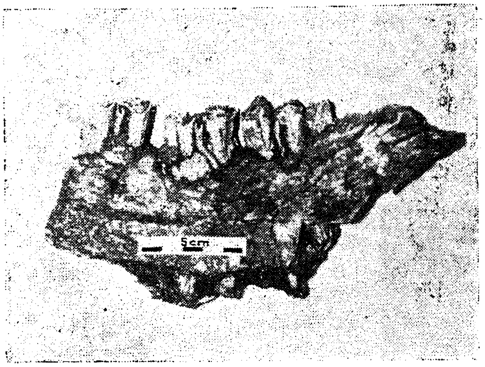

FIG. 1. Left ramus of mandible with 1st, 2nd and 3rd, molar teeth.

Comparative measurements of teeth of *Bos namadicus* from Susunia and of specimens from other locatities present in the G.S.I. Gallery, Indian Museum, are given in Table I.

TABLE I

Comparative measurements in millimetres of teeth of Bos namadicus *Falconer from Susunia and species from other localities*

	Length of the crown of lower 3rd molar	Width of the crown of lower 3rd molar	Length of the crown of lower 2nd molar	Width of the crown of lower 2nd molar	Length of the crown of lower 1st molar	Width of the crown of lower 1st molar
Specimen from Susunia. (left mandible)	46·3	16·2	35·0	15·7	31·6	15·5
Bos namadicus from Narbada alluvium Regd. No. F–62 (mandible)	46·1	16·8	30·0	16·5	25·8	16·0
Bos namadicus from Kathiawar Regd. No. F–68 (3rd lower molar)	46·0	17·5	..	..	..	..
Bos namadicus from Narbada alluvium Regd, No. F–66 (left mandible)	42·5+	16·0	30·0	16·0	..	..
Bos namadicus Type No. 18043 sp. No. K49/905 (mandible)	40·1	15·0	33·2	17·2	30·0	15·3

Discussion

The above measurements show that the 3rd molar teeth in our material have full resemblance, in length and width of the crown, with those in authentically identified specimens of *Bos namadicus* Falconer from Narbada alluvium and Kathiawar. The 2nd and 1st molar are nearly similar except that they are somewhat larger in our material.

It is certain that the piece of mandible with the molar teeth from Susunia is similar to those described for the fossil species *Bos namadicus* Falconer.

The fossil *Bos namadicus* has been reported from India (Pilgrim, 1939) as early as middle Pleistocene from Narbada alluvium, old alluvium of Penganga and Jamuna River and Billa Surgam cave of Kurnool. The age of the mandible from Susunia discussed here is estimated to be about 20,000 years old. It, therefore, would be the youngest among fossil specimens of this species earlier recorded from other areas in India and incidentally constitutes the first record of this species from Bengal in eastern India. Judging from the fact that one of us (S. Banerjee) has obtained some teeth of this species from Mochpal, Barasat, 24-Parganas District; about 30 Km. east of Calcutta along with partially petrified bones of *Bubalus palaeindicus* Falconer and some early pottery wares, a prehistoric sites ascribed to the Mauryan tradition period (*c* 2000 years) indicate that this species survived recently in the eastern India and it seems to have had a fairly wide distribution in the late tertiaries in the Indian peninsula.

Against this context we may have to revise our ideas about the identity of the voluminous bovine remains excavated from the prehistoric sites of the so called Indus Valley Civilisation at Harappa, Mohenjodaro, and other areas in India.

We are thankful to the Director, Zoological Survey of India, for the facilities and to Dr. P. C. Das Gupta, Director of Archaeology, Government of West Bengal, for the material of study. We are grateful to Shri M. V. A. Sastry, Director, Palaeontology Division, for the fossil collections and thanks are also due to Dr. K. K. Tiwari and Dr. B. Biswas, for critically going through the manuscript.

Zoological Survey of India,
8, Lindsay Street,
Calcutta 700 016, *June* 9, 1975.

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---

1. Pilgrim, G. E., "The Fossil Bovidae of India," *Palaeont. Indica* (*N.S.*), 1939, 26 (1), 318.

ARTERIOSCLEROSIS ASSOCIATED WITH PULMONARY OSSIFICATION IN ALPINE GOATS

The association of arteriosclerosis with pulmonary ossification is of rare occurrence in goats. However, this association in sheep was observed long ago in Argentina[4] and recently in Punjab in large number of Corriedale sheep[3]. In India a number of cardiovascular investigations have been carried out[2,5,6], but none of these indicated any such association suggesting the rarity of this combination in goats. The present communication deals with the study of 94 Alpine goat carcases subjected to necropsy and followed by routine histopathology. For detection of calcium salts von Kossa technique was employed.

At necropsy, unusual prominence of arota was observed in 5 cases. The vessel was hard to cut and on opening mineralized plaques were seen scattered over the entire intimal surface of the organ (Fig. 1), however, the extent of mineralization varied with the individual cases. In advanced cases the plaque formation resulted in appreciable narrowing of the lumen. Focal whitish spongy areas in the lungs produced grating sound while cutting, owing to mineral deposition in this organ too. The microscopic examination of aorta revealed focal or diffuse basophilic granular deposits in the tunica media. In extensively calcified regions the presence of osteoid tissue (Fig. 2), alongwith degenerative changes in the adjacent tissues were noticed. The myocardial muscles also suffered degenerative changes leading to necrosis. In two cases laying down of mineral salts accompanied with fibrous tissue proliferation in lungs resulted in marked thickening of interalveolar septa. Metaplastic leading to well marked areas of ossification characterized by the formation of osteoid cells in

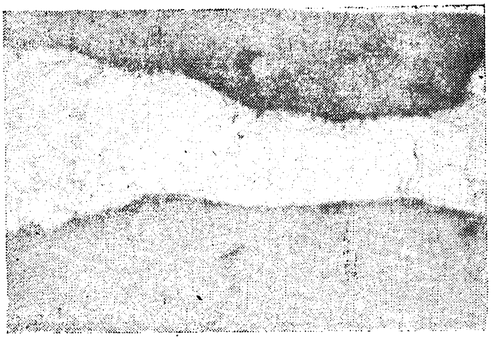

FIG. 1. Gross specimen of aorta showing mineralized plaques over the entire intimal surface.

the severely calcified regions of lungs were also noticed (Fig. 3). Calcification also affected the tunica media of some renal arteries. The calcified foci developed blackish coloration on von Kossa's staining.

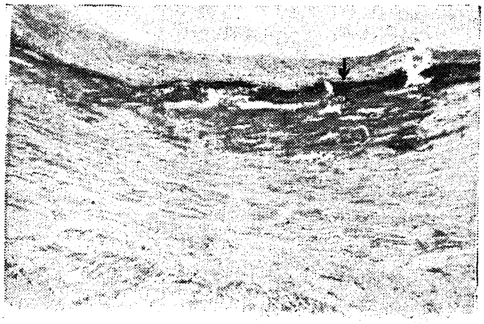

FIG. 2. Section of aorta showing severe calcification with few osteoid cells (arrow) in the tunica media. H.E. × 75.

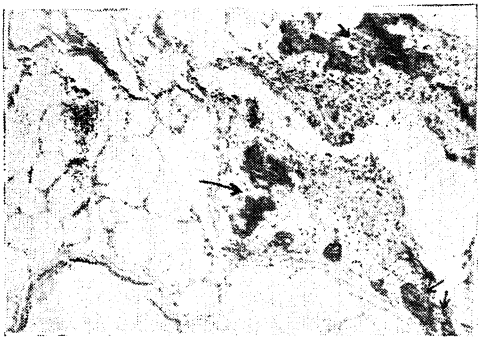

FIG. 3. Section of lung showing calcification and ossification (arrows) replacing considerable portion of pulmonary tissue. H.E. × 75.

In goats, the cases of arteriosclerosis[6] as well as pulmonary ossification[1] in isolation are on record but the information about the association of both the conditions in goats as observed during the present study is lacking in the available literature. In India it seems to be the first report of its kind affecting goats, however arteriosclerosis in sheep accompanied with metaplastic changes leading to ossification in lungs and some other organs has recently been recorded in Punjab[3]. The estiology of this condition is still obscure but mineral imbalance seems to be the likely-possibility.

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---

1. Charan, K., Iyer, P. K. R. and Kaushal, B. S., *Indian J. Anim. Sci.*, 1972, 42, 824.
2. Chattopadhyay, S. K. and Sharma, R. M., *Ibid.*, 1972, 42, 698.
3. Hothi, D. S. *Ph.D. Thesis*, Punjab Agricultural University, 1974.
4. Lignieres, J., *Newsom's Sheep Disease*, 3rd Ed. Williams and Wilkins, Co., Baltimore, 1912, p. 351.
5. Pande, P. G., Sharma, G. L. and Bhalla, N. P., *Proc. 48th Indian Sci. Cong.*, 1961, Part IV, 16.
6. Prasad, M. C., Rajya, B. S. and Mohanty, C. C., *Exptl. Mol. Path.*, 1972, 17, 14.

STUDIES ON SALIVARY ENZYMES OF *RAGMUS IMPORTUNITAS* DISTANT (HEMIPTERA: MIRIDAE)

INSECTS deposit salivary secretion in or on plants when feeding which significantly influence the physiology of the surviving tissues. Some of the secretions result in phytotoxemia including galls in plants. Previous reviews on salivary secretions, particularly that of Hemiptera, were those of Carter[1] and Miles[2]. The mirid, *Ragmus importunitas* Distant causes serious damage to sunnhemp (*Crotalaria juncea* L.) resulting in chlorotic leaves, cessation of growth and considerable loss of seeds[3]. Therefore attempts have been made to identify the enzymes present in the salivary glands of *R. importunitas* so as to understand the physiology of feeding and the results are presented below.

The salivary glands were dissected from freshly collected adults and nymphs of *R. importunitas* and the salivary extract was prepared, as per the method of Hori[4], by homogenizing the contents in distilled water and centrifuging at 26,000 g for 5 minutes. The clear supernatant was used as the enzyme preparation. One ml of the solution contained extract from 5 to 10 pairs of salivary glands. The enzymes in the salivary glands were detected as per the methods described for each, *viz.*, pectic enzymes[5], macerating enzymes[6], cellulase[7], amylase and protease[8], other carbohydrases[9], lipase[10] and polyphenol oxidase[11].

The salivary glands of *R. importunitas* were found to contain pectic enzymes, macerating enzyme(s), cellulase, amylase, protease and polyphenol oxidase. The pectic enzymes were identified only in the posterior lobe of the principal gland. The enzyme activity was persistent even upto 10 days after dissection. Macerating enzyme capable of digesting potato tissue was evident in the salivary apparatus. Complete maceration of potato disc of 10 mm diameter and 30 μ thick occurred in 5 hr per ml of the enzyme solution containing 10 pairs of salivary glands. The presence of cellulase (Cx) is recorded for the first time in the salivary apparatus of Miridae though this has previously been reported in the salivary glands of aphids[12]. Mean relative activities (RA) of 70, 45 and 38 were observed in the case of the salivary glands (10 pairs/ml) of females, males and fifth instar nymphs respectively. The salivary glands of *R. importunitas* were found to contain the enzyme amylase as revealed by the clear spots on the starch-agar coated slide following staining with dilute iodine. Only the posterior lobe of the principal gland showed amylase activity. The activity of other carbohydrases, *viz.,* maltase, invertase, glucosidase and galactosidase was not noticeable in the salivary glands. Proteolytic enzyme activity was evident only in the posterior lobe of the principal gland. Polyphenol oxidase activity was detected only in the accessory glands. No lipase activity could be detected in the salivary glands.

The activity of the salivary enzymes found in *R. importunitas* in comparison with the reported levels of other mirid bugs is given in Table I.

TABLE I

Salivary enzymes activity of R. importunitas *in comparison with other mirid bugs*

Enzymes	*R. importunitas*	*Lygus hesperus* Knigh. (Strong and Kruitwagen[5])	*L. disponsi* Linnavuori (Hori[4, 13])
Endo PG	350 RA	800 RA	..
Macerating enzyme	5 hr	48·4% digestion in 6 hr	..
Cellulase	70 RA	..	..
Amylase	530 μg glucose/ ml/24 hr; at pH 7·0	..	1·75 mg glucose/ ml/24 hr at pH 7·0
Protease	280 μg tyrosine/ ml/24 hr at pH 7·0	..	220 μg tyrosine/ ml/24 hr at pH 7·0

RA = 1000 × reciprocal of the time in min necessary for 50% reduction in viscosity of the substrate.

Endo PG = NaPP substrate at pH 5.

Cellulase = Carboxy methyl cellulose at pH 5.

Macerating enzyme = Time taken in min to macerate potato discs of 10 mm dia and 30μ thick.

The presence of pectolytic, macerating and cellulolytic enzymes in the salivary glands of *R. importunitas* is interesting and assumes much significance. Both the pectic and cellulolytic enzymes are considered to be the most important biochemical offensive weapons in the host cell-wall degradation[14, 15].

The presence of these enzymes in the salivary apparatus of *R. importunitas* may be responsible for the maceration and dissolution of plant cells facilitating penetration of the tissues by the stylets and for removal of the cell contents. The amylase and protease in the salivary glands may take part in the external digestion and supply of nutrients to the insect. The presence of polyphenol oxidase in the accessory glands of the salivary apparatus is important since this may be ejected by the insect at times during feeding and is considered by Miles[16] as a counter to overcome the host plant's defensive mechanism by speeding up the process of oxidation of phenolics to non-toxic end products. Hence the presence of these enzymes in the salivary apparatus may particularly be responsible for the severe phytotoxemic effects for which this group of insects are so notorious.

This work forms part of the Ph.D. thesis and the author wishes his sincere thanks to Dr. T. R. Subramaniam, Head, Department of Entomology, Tamil Nadu Agricultural University, Coimbatore, for his guidance.

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Madurai, *October* 28, 1975.

---

1. Carter, W., *Insects in Relation to Plant Disease,* Wiley, Interscience Publications, New York, 1973, p. 759.
2. Miles, P. W., *Adv. Insect Physiol.,* 1972, **9**, 183.
3. Gopalan, M. and Basheer, M., *Madras Agric. J.,* 1966, **53**, 22.
4. Hori, K., *J. Insect Physiol.,* 1970, **16**, 373.
5. Strong, F. E. and Kruitwagen, E. C., *Ibid.,* 1968, **14**, 1113.
6. Brown, W.. *Ann. Bot.,* 1915, **29**, 313.
7. Hancock, J. G., Millar, R. L. and Lorbeer, J. W., *Phytopathology,* 1964, **54**, 928.
8. Pickford, O. E. and Dorris, F., *Science,* 1934, **80**, 317.
9. Saxena, K. N. and Bhatnagar, P., *Proc. Nat. Insect. Sci. India,* 1958, **24 B**, 245.
10. Bier, M., *Methods in Enzymology,* I Ed. by Colowick, S. P. and Kaplan, N.O., Academic Press, New York, 1955, p. 627.
11. Miles, P. W., *J. Insect Physiol.,* 1964, **10**, 121.
12. Adams, J. B. and Drew, M. E., *Can. J. Zool.,* 1965, **43**, 489.
13. Hori, K., *Res. Bull. Obihiro. Univ.,* 1970, **6**, 318.
14. Bateman, D. F. and Millar, R. L., *Ann. Rev. Phytopathol.,* 1966, **4**, 119.
15. Goodman, R. N., Kiraly, Z. and Zaitlin, M., *The Biochemistry and Physiology of Infectious Plant Diseases,* D. Van Nostrand Company, Inc., 1967, p. 354.
16. Miles, P. W., *Ent. exp. appl.,* 1969, **12**, 736.

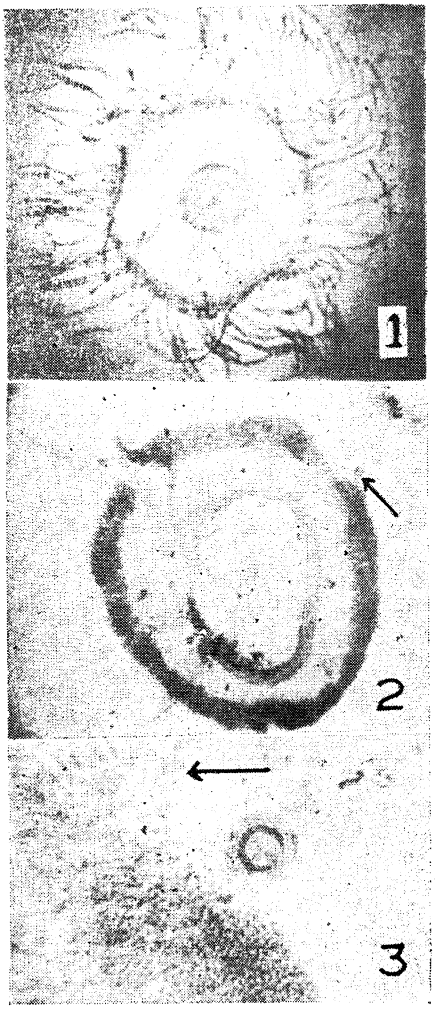
MANSARIELLA LACUSTRIS, GEN. *ET* SP. NOV., A NEW FRESHWATER MEDUSA FROM JAMMU, INDIA

THE reports[1-3] on the occurrence of freshwater medusae refer to the collections made from rivers and lakes from south of Vindhyas. A solitary report[4] of the occurrence of medusae (*Moerisia gangetica*) from Gangetic water system, thus appeared highly significant. The freshwater habit of this species was controverted and the form was believed to be an "estuarine species with all its nearest relatives either lacustrine, marine or pelagic[5]".

The present form, named here *Mansariella lacustris* gen. *et* sp. nov., was found to occur in large numbers during September and October in Mansar Lake, Jammu, India. It is a subtropical, isolated freshwater lake with apparently no escapes. Seemingly it is a spring-fed lake aided further by monsoons and local precipitation. In situation, it is sub-Shivalik at a height of 700 m above sea level, with a near oval to circular contour, reaching a diameter of around 400 m in its most swollen condition. The different physico-chemical analyses of the water are being carried out.

The medusae (Fig. 1) are umbrella-shaped, each attaining a maximum diameter of 15 mm at its sub-umbrella margin. The margin is beset with highly extensile tentacles (reaching over 20 mm in maximum stretched condition), numbering nearly 160 in total (around 40 in each quadrant). The tentacles are arranged in a single row and do not show any terminal sucker or an adhesive pad. In the living condition the bases of tentacles are swollen, with each swelling containing a statocyst. The mouth is circular 5 mm dia, when open Manubrium is short and opens into a circular gastric cavity from where, emerge 4 radial canals at right angles to one another. No gonads, however, were seen in any specimen examined. Young medusoid forms appeared to bud off (Figs. 2 and 3) from the gastric region of the bigger forms. Each young medusa bears a circlet of 16 closely set tentacular buds all along the rim of the umbrella. But for size, number and simplicity in its tentacles, each young medusa shows all identifiable structures of the adult.

The Indian peninsular forms (*Limnocnida, Limnocodium* and *Microhydra*) differ markedly from the present form in the number and arrangement of the tentacles and the comparatively higher mouth-umbrellar diameter ratios. Even the gangetic form, *M. gangetica,* which Jones[6] seems to have overlooked, stands no comparison with the present form, in the number of tentacles, the shape of the umbrella or any other physical parameter.



FIGS. 1–3. Fig. 1. Entire medusa as seen ventrally (20 ×). Fig. 2. A young medusa (arrow) budding from mother. The mouth aperture is fully open to show oval outline (40 ×). Fig. 3. The young medusa in relation to the radial canal (arrow) of the mother (200 ×).

Nevertheless, the reports of the occurrence of *Craspedacusta* in very unpredictable manner, in water bodies of varying nature, all over the world[5], appears interesting and significant. But a comparison of *M. lacustris* with *Craspedacusta sowerbeyi* from India reveals differences in features such as (*i*) umbrellar depth, (*ii*) oral aperture, (*iii*) tentacular number, (*iv*) tentacular length and (*v*) the mode of asexual reproduction.

The present form is, therefore, morphologically distinct from all other known freshwater medusae, The differences, being taxonomically significant, warrant a separate generic and specific treatment for the form described. It is, thus, proposed to be named as *Mansariella lacustris* (the generic name after the name of the lake from where it was discovered and the specific name after the nature of the freshwater of the lake). How these forms came to inhabit such isolated waters as Mansar lake in Jammu, may remain a matter of speculation.

Holotype deposited in the museum, Department of Biosciences, University of Jammu.

Department of Biosciences, University of Jammu, Jammu 180 001 (J & K), *December* 23, 1974.

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M. K. JYOTI.

1. Hora, S. L., *J. Bomb. Nat. Hist. Soc.*, 1926, 31, 449.
2. Rao, S. H., *Ibid.*, 1932, 36, 210.
3. Ramakrishna, P. A., Bhimachar, B. S. and Subramaniam, M. K., *Ibid.*, 1950, 49, 319.
4. Kramp, P. L., *Rec. Ind. Mus.*, 1958, 53, 399.
5. Joshi, M. V. and Tonapai, G. T., *Curr. Sci.*, 1965, 34, 665.
6. Jones, S., *J. Bomb. Nat. Hist. Soc.*, 1958, 53, 799.

TWO NEW RECORDS OF COELOMYCETOUS FUNGI FROM INDIA

DURING her mycological survey around Poona, the writer collected two coelomycetous fungi one of which was found to be a new species and the other a new generic record to Indian Fungi, detailed descriptions of which are presented below.

1. *Amerosporium* Speg.

A non-ostiolate pycnidial fungus with dark setae, and producing one celled, hyaline conidia on phialophores was collected on roots of *Ficus benghalensis* and was identified as belonging to form-genus *Amerosporium* (Sphaeropsidales). Two species of this genus have been so far reported from India, one by Gupta (1967) as *A. applanatum* Berg. *et* Curt. isolated from rhizosphere of *Withania somnifera* Linn. and the other by Wehmeyer (1964) as *A. virida* Wehm. growing on *Duthiea bromodes* Hack. Comparison of the writer's collection with these two Indian collections and the type species (*viz., A. polynematoides* Speg.) proved it to be new to science and hence is described here as *A. indicum* sp. nov. with Latin diagnosis.

Amerosporium indicum sp. nov. (Fig. 1).

Pycnidia nigra, non-ostiolata, errumpentia vel superficialia, setosa, 240–320 × 180–240 μ. Setae nigrae vel fusc brunneae, ad apicem pallescens. Parce septatae 200–300 × 4–6 μ. Conidiophora hyalina, phialophora. Conidia phialospora, unicellularia, hyalina, massa conidiarum fusc virido-olivacea; conidia elliptica ad basim truncata, ad apicem acuta, non-guttulata, 8–12 × 3–4 μ.

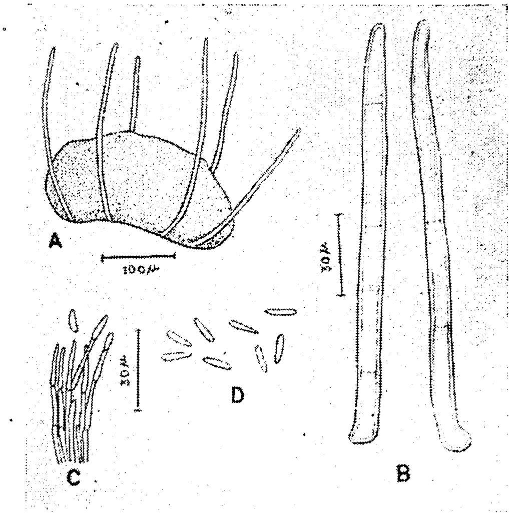

FIG. 1. *Amerosporium indicum* sp. nov. A, Pycnidium ; B, Setae ; C. Conidiophores ; D, Conidia.

Habit : On exposed roots of *Ficus benghalensis* collected at Jejuri (Dist. Poona) dt. 30–3–1975 by Mrs. Alaka Pande, deposited at Ajekar Mycological Herbarium of M.A.C.S. under AMH No. 2548.

Remarks : The fungus was isolated in pure culture and produced pinkish, slow-growing colonies on M_2 Agar with numerous hard, black pycnidia concentrated towards the center. The pycnidia and conidia were found to have the same dimensions as on natural host.

2. *Discula* Sacc.

A leaf spotting acervulate fungus was collected on leaves of *Olax wightiana* during late winter, which at first sight appeared to be a species of *Gloeosporium*. Literature review revealed that such fungi with intra- or sub-epidermal acervuli, having phialophores producing one celled, oval to elliptical hyaline phialospores were placed by Arx (1970) and Morgan–Jones *et al.* (1972) Saccardo's genus *Discula*. Comparison with the type species revealed that the present collection agreed with the description of the type species and hence is identified as *Discula platani* (Peck.) Sacc. Since this is a new generic record to India, a brief description is provided.

Discula platani (Peck.) Sacc.

Infection spots foliicular, necrotic ; upto 2 cm in diam. Acervuli intra-epidermal or sub-epidermal, arranged in concentric incomplete rings on the spots; 160–200 × 48–60 μ. Basal stroma subhyaline, giving rise to conidiophores. Conidiophores phialidic, in basal palisade, simple or branched, hyaline; conidiophores may show one or two annellations. Conidia phialosporous, elliptical guttulate, 12–15 × 4·5–6 μ.

Habit: On living leaves of *Olax wightiana* Wall. collected at Karnala Fort (elevation 1,600 ft. above m.s.l.) (Dist. Kolaba) dt. 26–2–1975. Leg. Mrs. Alaka Pande, deposited in Ajrekar Mycological Herbarium at M.A.C.S. under AMH No. 2459.

The writer is thankful to Prof. M. N. Kamat, for guidance and to the Director, M.A.C.S., for facilities.

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Law College Road,
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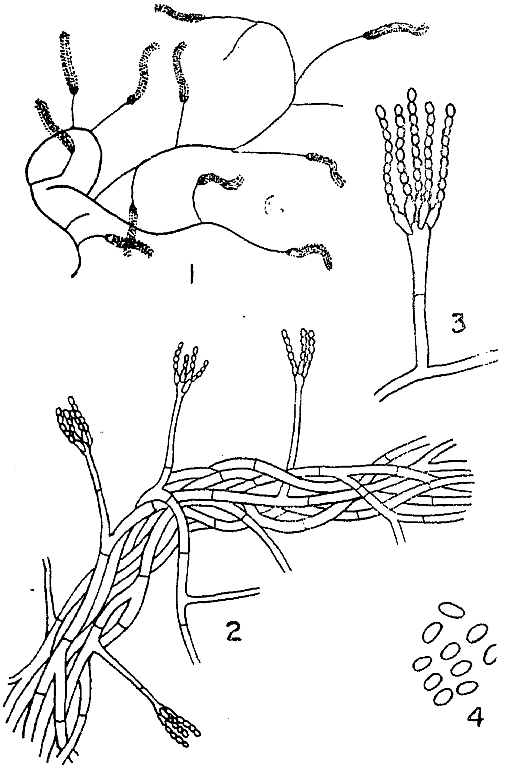
1. Arx, J. A. von, "A revision of the fungi classified as *Gloeosporium*," *Bibliotheca Mycologica*, 1970, Band 24, 203 (English Translation).
2. Gupta, P. C., *Curr. Sci.*, 1967, 36, 415.
3. Morgan Jones, G., Nag Raj, T. R. and Bryce Kendrick, *Icones generum Coelomycetum—V.* Univ. Waterloo Biol. Ser., 1972, p. 52.
4. Wehmeyer, L. E., *Mycologia*, 1964, 56, 29.

PENICILLIUM BREVISSIMUM SPEC. NOV. FROM INDIAN SOILS

THE paper reports a new species of *Penicillium*, *P. brevissimum* of *Penicillium decumbens* series Raper and Thom[2]. It is characterized by the production of green, strongly funiculose colonies bearing somewhat short conidiophores with loosely arranged sterigmata which are fewer per verticil. All colours mentioned in this paper are from the Horticultural Colour Chart of the British Colour Council[1]. Raper (personal communication) is of the view that "it belongs to the *Penicillium decumbens* series but does not fit exactly with any of the species described in the manual".

Penicillium brevissimum spec. nov. (Figs. 1–4):

Coloniae in agaro ordeaceo lente crescentes, "Sage Green" (000861/2) fortiter funiculosae, constantes texture basali tenaci coriacea, marginibus irregularibus praedita, alte sporulante in centro. Conidiophora brevissima 11·2–80·0 μ × 3·2–5·4 μ, parietibus levibus ornata, emergentia hyphis repentibus at intertextis aeries et funibus hypharum, desinentia in bene definitos apices vasicularis 8·0–10·5 μ. Penicillia stricte monoverticillata. Sterimata 3–6 laxe disposita ad apices vesiculares conidiophororum 6·9–8·4 μ × 2·8–3·8 μ. Conidia insidentia columnis laxe et saepe varie flexis, elliptica vel cylindrica, levia, patilum crasse parietata, in mass a viridia 3·0–3·8 μ × 1·5–3·0 μ.



FIGS. 1–4. *Penicillium brevissimum* spec. nov. on Malt-extract-agar. Fig. 1. Showing shortly branched conidiophores, × 150. Fig. 2. Conidiophores arising from basal funiculose hyphae, × 350. Fig. 3. Conidiophore showing strictly monoverticillate penicillus, × 375. Fig. 4. Conidia, × 510.

Colonies on malt-extract agar slow growing attaining a diameter of about 2·5 cm in 12 days, "Sage Green". (000861/2) consisting of a tough leathery basal felt, with thin irregular whitish margins, heavily sporulating in centre, reverse colourless to pale, exudates entirely lacking. Conidiophores 11·2–80·0 μ × 3·2–5·4 μ, smooth walled, arising from trailing and interlacing aerial hyphae ending in a more or less well defined vesicular apices 8·10–10·5 μ in diameter. Penicilli strictly monoverticillate sterigmata 3–6, loosely arranged on vesicular apices 6·9–8·4 μ × 2·8–3·8 μ. Conidia borne in loose parallel columns often variously bent, elliptical to cylindrical, 3·0–3·8 μ × 1·5–3·0 μ, smooth slightly heavy walled, green in mass.

Colonies on Czapek-Dox agar fairly slow growing, tough, prominantly wrinkeld, sporulating only in central areas bearing very dimunitive conidial heads.

Isolated from rhizosphere soil of *Brassica campestris* var. *toria*, Lucknow, India. A transfer from the type culture deposited in the Department of Bacteriology, University of Wisconsin, under the reference number WB 5418 and a subculture from this deposited in Centraalbureae Voor, Schimmelcultures—Baarn (Netherland), as CSB 763·68.

Thanks are due to Professor Kenneth, B. Raper, Dr. D. B. Prest and Dr. Amelia C. Stolk, for valuable comments.

Department of Botany,
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Lucknow, *July* 21, 1975.

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1. *Horticultural Colour Chart*, Issued by the British colour Council in collaboration with the Royal Horticultural Society, Henry Stone and Son Ltd., Banbury, Great Britain, 1938.
2. Raper, K. B. and Thom, C., *Manual of Penicillia*, The Williams and Wilkins Co., Baltimore, 1949.

EFFECT OF PRETREATMENT OF SEEDS AND WATER STRESS ON TRANSPIRATION RATE IN BAJRA (*PENNISETUM TYPHOIDES* STAPF AND HUBB.)

TRANSPIRATION rate in plants provide an index for drought resistance. Recently anti-transpirants like PMA have been employed for reducing the transpiration in many species with satisfactory results. Pre-treatment of seeds with specified growth substances have also been suggested, the object being to induce certain degree of tolerance to drought. In all these cases it is generally accepted that the chemicals enhance the water retaining capacity of the protoplasm. The present work was initiated with the object of testing the efficacy of the growth regulators, CCC (5 ppm), resistine (10 ppm) kinetin (5 ppm) and seed hardening, in inducing tolerance to drought in *Pennisetum typhoides* (HB 3). Water stress was created to maintain 40, 50 and 70% water holding capacity. This was done using water holding capacity cups initially and thereafter, the respective levels were maintained in weight basis. Transpiration rate was measured by maintaining the water holding capacity of each pot until harvest stage.

Genkel and Badanova[1], observed that hardened plants showed a higher rate of transpiration while Milthrope[2], recorded that hardening increased the water holding capacity of protoplasm. According to Nunes[3], kinetin (50 ppm) had no effect on transpiration rates of wheat plants. Salim and Todd[4] stated that pre-sowing seed treatment seemed to induce higher overall water holding capacity in leaves, compared to check.

From the results (Table I) it is evident that all the treatments reduced the transpiration rate in the chosen species. In respect of each treatment, transpiration rate increased with higher water holding capacity, the rate of increase being significantly different between 40–50 and 50–70% water holding capacity (WHC). From the general trend it seemed possible that there was scope for reduction in WHC to arrive at an optimum transpiration rate. The capacity to tolerate drought has been

TABLE I

The effect of Pre-treatment on transpiration rate of Pennisetum typhoides

	40 per cent.				50 per cent.						70 per cent.			Water holding capacity (WHC) interval (days)	
	20	40	60	80	95	20	40	60	80	95	20	40	60	80	95
	(Transpiration rate in cubic centimeters per day)														
Control	90	100	160	100	100	100	180	180	120	100	110	200	200	200	140
Seed hardening	60	90	120	90	90	90	100	100	100	100	100	140	150	100	110
CCC 5 ppm	40	50	60	30	40	70	80	90	60	60	90	90	100	90	70
Resistance 10 ppm	40	40	40	40	40	60	70	80	80	60	80	90	90	90	90
Kinetin 5 ppm	50	60	50	40	20	60	70	70	60	70	80	80	90	90	80

(i) Stages: C.D. 10·43 (P=0·005)
(ii) WHC: C.D. 8·08 (P=0·05)
(iii) Treatments: C.D. 80·76 (P=0·05)

Conclusion: 1. Stages (days) 60 40 80 95 20
2. WHC: 70%, 50%, 40%
3. Treatments; Control, Seed hardening, CCC 5 ppm, Resistine—10 ppm, Kinetin 5 ppm

more, particularly after 60th day in kinetin treatment. It was also noted that all the treatments were effective in inducing tolerance to drought. Kinetin, resistine and CCC were equally effective and better than hardening.

Between the stages, the lowest rates of transpiration was in the initial stage (20 days) while the highest rates were on the 60th day. The results were indicative of the fact that the chosen pretreatments can effectively reduce the transpiration rates, although the actual mechanism needs to be investigated. Genkel and Badanova[1], recorded higher rate of transpiration in hardened plants. Genkel[5], attributed this situation to more intense transpiration and more water deficit which were due to more bound water and greater xeromorphic structure.

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1. Genkel, P. A. and Badanova, K. A., *Fd. Crop. Abstr.*, 1959, 15, 93.
2. Milthrope, F. L., *Ibid.*, 1959, 12, 1.
3. Nunes, M. A., *Ibid.*, 1967, 22, 79.
4. Salim, M. H. and Todd, G. W., *Agron. J.*, 1968, 60, 179.
5. Genkel, P. A. (Henkel, P. A.), *Proc. Madrid Symp. Paris. UNESCO*, 1961, p. 167.

INSOLUBLE POLYSACCHARIDES IN *FOENICULUM* OVULES

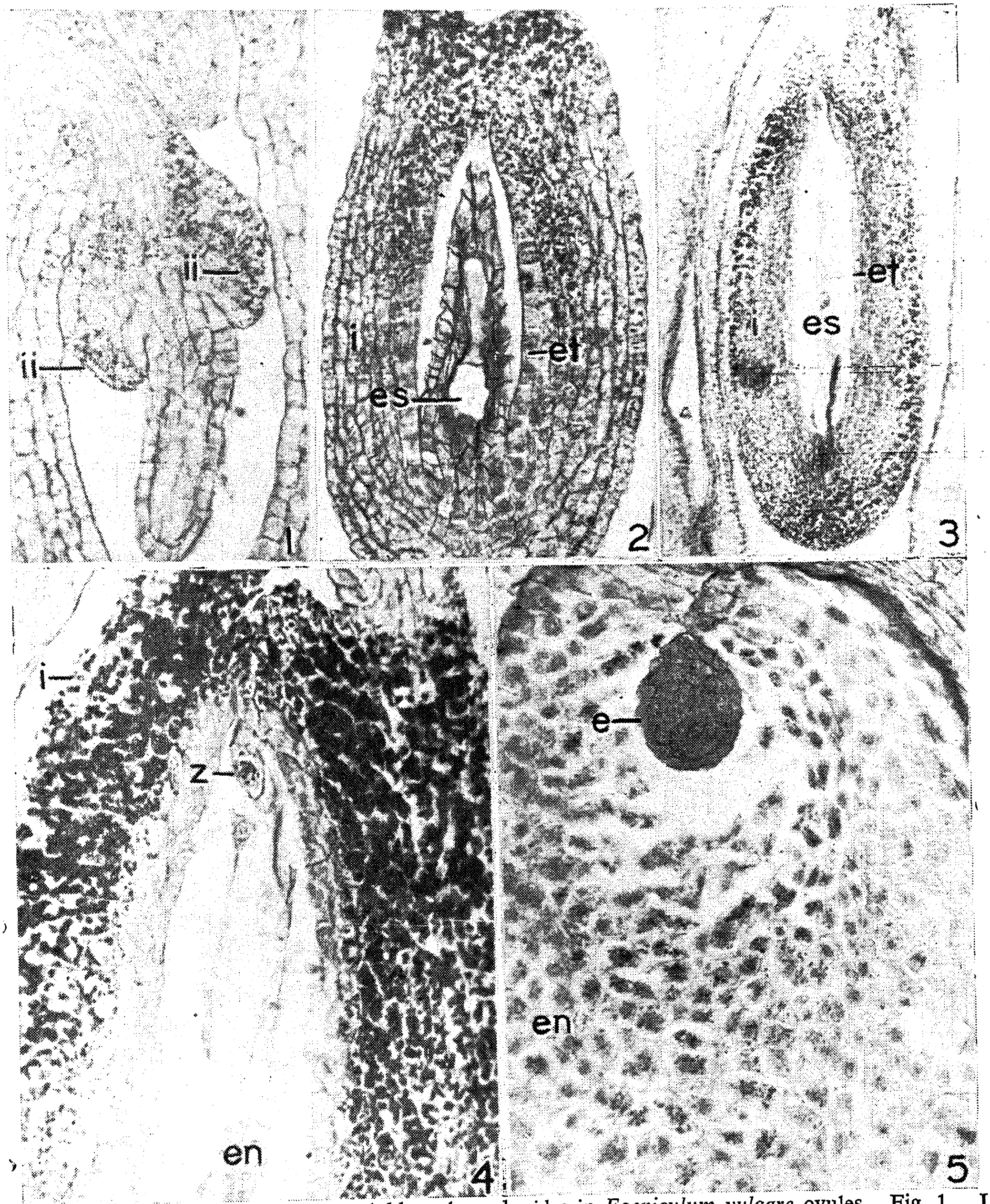
IN the Umbelliferae, the cells of inner epidermis of ovular integument develop into endothelium-like cells, but the presence of a functional endothelium has been doubted earlier[1]. Hence, qualitative histochemical studies of ovules have been undertaken for the *in situ* localization of insoluble polysaccharides, proteins, nucleic acids, histones, etc. The present communication reports only the distribution of insoluble polysaccharides of total carbohydrates in developing ovules of fennel.

Flower buds at different developmental stages were fixed in formalin-acetic-alcohol. The usual paraffin method was followed for microtomy. Sections of thickness between 6–10 microns were cut. One set of slides was stained with safranin-fast green and the other was treated with periodic acid Schiff's (PAS) reaction[2]. Controls were run by (i) omitting the periodic acid treatment and (ii) by acetylation[3]. In *Foeniculum vulgare,* the ovules are unitegmic and tenuinucellate[4]. The endothelium differentiates at 2-nucleate stage of embryo sac when its cells appear slightly radially elongated. However, the cytoplasm is not dense enough to recognise this layer distinctly. At 4-nucleate stage, the cells get further elongated and persist up to 8-nucleate stage of embryo sac. It remains uniseriate all through and starts disintegrating after fertilization. Thus, the integumentary tapetum has a short life span.

Total insoluble polysaccharides are uniformly distributed in an undifferentiated ovule. As the archesporial cell is organised in the nucellus, its cytoplasm develops light PAS-positive tinge. Concurrently, the PAS-positive grains start appearing in the integumentary initials and also at the chalazal end of the ovule (Fig. 1). At 2-nucleate embryo sac stage, polysaccharide grains are concentrated more at the micropylar end of the ovule, 3 or 4 cell layers adjoining the endothelium, and the outer epidermal cells of the integument, but comparatively fewer in the endothelium (Fig. 2). Later, as the endothelial cells elongate, their cytoplasm as well as walls stain equally well like the rest of the integumentary cells. Simultaneously, at the 4-nucleate stage, in the micropylar end of the integument the density of PAS-positive grains declines.

The ovule, on the whole, becomes more PAS-positive at organised embryo sac stage. Uniform intense stain for polysaccharides appears in the cytoplasm of endothelium and adjoining 4–6 layers of cells, but they lack PAS-positive grains altogether. However, the polysaccharides accumulate in the form of grains in the outer peripheral cell layers of the integument, excepting a few cell layers on the raphe side which persist in the mature seed (Fig. 3). After fertilization the endothelial cells start breaking down but the zygote shows accumulation of polysaccharide grains. The PAS-positive grains increase remarkably in the peripheral cell layers of integument. The density of these grains decreases gradually centripetally (Fig. 4). Concomitantly, the inner cell layers of integument, adjacent to embryo sac, become devoid of the grains but their cytoplasm stains dark with PAS-reaction. Thus, a clear centrifugal gradient is set up in the integumentary cells, indicating the utilization of polysaccharides by the developing endosperm.

The integument gets depleted off of its polysaccharides at globular embryo stage. The cellular endosperm develops a high polysaccharide content which is concentrated more in its micropylar half. Some of the cells of endosperm in the vicinity of embryo start breaking down, and their cytoplasm stains darker. This is accompanied by a gradual increase of polysaccharides in the globular embryo (Fig. 5). Subsequently, the grains in the endosperm cells disappear but the cytoplasm assumes uniformly dense PAS-positive stain. Concomitantly, the embryo acquires high concentration of PAS-positive material, indicating the utilization of polysaccharides from its surrounding tissues.



FIGS. 1–5. Distribution of insoluble polysaccharides in *Foeniculum vulgare* ovules. Fig. 1. L.s. ovule at archesporial cell stage, × 312. Fig. 2. Same, at 2-nucleate embryo sac stage, × 371. Fig. 3. Same, at organised embryo sac stage. The PAS-positive grains are deposited at micropylar and chalazal ends, × 182. Fig. 4. Same, zygote showing PAS-positive grains. Note the accumulation of grains in the peripheral integumentary cell layers, whereas inner cell layers lack the grains completely, × 304. Fig. 5. Same, showing the globular embryo rich in polysaccharides; the endosperm cells in the vicinity of embryo are breaking down.

(*e*, embryo; *en*, endosperm; *es*. embryo sac; *et*, endothelium; *i*, integument; *ii*, integumentary initials; *z*, zygote.)

Thus, the present histochemical observations indicate that at least in *Foeniculum vulgare*, the endothelium is short lived. The distribution pattern of insoluble polysaccharides in endothelium shows that it does not differ significantly from other adjoining integumentary tissues and on the whole it is probably not very active. Nevertheless, it is evident that the insoluble polysaccharides, initially deposited in the integument are used up by the developing embryo through the intermediate nourishing tissue, the endosperm.

We are thankful to Dr. (Miss) Kanan Nanda and Miss V. M. Rajeswari, for helpful discussions. One of us (S. A.) acknowledges the financial assistance of the University Grants Commission.

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1. Gupta, Shrish C., *Ph.D. Thesis*, Delhi Univ., 1960.
2. Jensen, W. A., *Botanical Histochemistry*, W. H. Freeman and Co., San Francisco, 1962.
3. Barka, T. and Anderson, P. J., *Histochemistry : Theory, Practice and Bibliography*, Harper and Row Publishers, Inc., New York, 1965.
4. Gupta, Shrish C., *Phytomorphology*, 1964, 14, 530.

SHORT SCIENTIFIC NOTES

Inheritance of Anthocyanin Pigment in Linseed (*Linum usitatissimum* L.) Seedlings

Development of anthocyanin pigment was marked in several varieties of linseed seedlings. Varieties Neelum and EC. 77918 with pink seedlings in crosses with green seedling types, EC. 41599, EC. 1458 and EC. 12351 showed the monogenic dominance of pink pigmentation in seedlings. No segregation occurred when varieties within the same pigment groups were mated indicating the presence of the same gene for anthocyanin pigment. Pink seedling colour of Neelum—a released cultivar—can be used as a genetic marker in the purification of the variety as well as in the hybridization programme. These results have not been reported so far in linseed.

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A New Leaf-Spot Disease of *Capparis horrida* L.

During the course of an investigation on plant pathogenic fungi of District Jaunpur, infected leaves of *Capparis horrida* showing the presence of blackish patches on their surface, were collected during June–July, 1974.

Microscopic examination of the material revealed the presence of *pycnia*, which were densely scattered and embedded in the upper and lower surfaces of the leaf lamina. The pycnia were dark-brown to black in colour, sub-epidermal, globose to subglobose 162–215 μ in diameter, thick (30–60 μ) walled, and composed of 5–7 layers of elongated polyhedral brown cells; ostiolate, ostiole 20–40 μ wide and usually 45 μ in length; conidiophores hyaline, minute, 7·5–10·5 μ in length and ·75–1 μ in thickeness. Conidia very minute, densely filling the cavity of the pycnidium.

The fungus has been identified as *Leptodothiorella* sp., the spermatial state of *Guignardia creberrima* Syd. by Dr. Punithalingam of C.M.I., Kew. Based on the perfect state the fungus may be named as *Leptodothiorella creberrima.*

The specimen has been deposited in C.M.I., Kew, at No. S 2 (IMI, 194875).

The fungus is a new record for India, and *Capparis horrida* is a new host record for this fungus.

The authors are thankful to Dr. Y. B. Singh, Principal, St. Andrew's College, Gorakhpur, for providing the facilities for the work and to Dr. Punithalingam, for providing the identity of the fungus. Thanks are also due to Sri S. K. Prabhuji, of our Department, for his kind help.

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October 12, 1975.

Responses of Seeds and Isolated Embryos of *Codiaeum variegatum* Blume var. 'Karna' *in vitro*

The potentialities of endosperm for continuous growth and morphogenesis have been well established species. This note describes the *in vitro* in several responses of decoated seeds and isolated embryos of *Codiaeum variegatum* Blume var. 'Karna'. Following the aseptic technique, the cultures were raised on a modified Whites' medium with 2% sucrose (BM) alone or along with various growth adjuvants and maintained under controlled conditions of temperature, light and humidity.

The decoated seeds containing a massive endosperm and fully differentiated embryo germinated to give rise to normal seedlings on BM, and on BM + CM (10%) alone or along with 2, 4-D (1 ppm). But on BM + CM (10%) + CH (500 ppm) + 2, 4-D (1 ppm), the seeds enlarged slightly and the endosperm started proliferating. In about six weeks, the endosperm produced a friable mass of yellowish-white callus. The latter was subculturable and comprised uninucleate cells of diverse shapes and sizes with dense cytoplasm and plenty of starch grains.

The fully differentiated isolated embryos, on a similar medium as above, developed proliferations from the radicular region and the cotyledonary leaves yielding a mass of yellowish-white, less compact tissue in five weeks. This tissue subsequently differentiated several roots which were normal and were studded with numerous root hairs. In addition the callus also contained several pockets of vascular tissues, and multicellular structures which often simulated stages in normal embryogeny.

We thank Professor M. S. Chennaveeraiah, Head of the Department of Botany, Karnatak University, for help and encouragement.

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September 26, 1975.

A Note on the Inheritance of Pigmentation in the Coleoptilar Leaf of Pearl Millet (*Pennisetum typhoides* S & H)

A number of workers have given the inheritance of qualitative characters in pearl millet (Singh *et al.*, 1967, 1968 and 1969; Gill, 1969 and Yadav, 1971), but no information is available on the inheritance of pigmentation in this leaf.

The material for the present study consisted of inbred lines (BS 1 and BS 3 with purple pigmentation and BS 2 with normal green colour in the leaf), their F_1's F_2's and both the back crosses were sown at the R.B.S. College farm in a randomised block design with three replications during the crop season of 1970.

The mode of inheritance of this character was studied in the progenies of F_1, F_2 and the back crosses. The data showed the dominance of anthocyanin pigmentation in the F_1 generation. Details of the segregation in the F_2 and back-cross generation is presented in Table I.

TABLE I

Inheritance of pigmentation in the coleoptilar leaf of pearl millet

Cross	Segregation in		Chi-square in		P-value in	
	F_2	test-cross	F_2	test-cross	F_2	test-cross
BS 1 × BS 2	85P 33NP	56P 62NP	0·269	0·994	70·50	50·30
BS 3 × BS 2	67P 21NP	41P 38NP	0·590	0·162	50–30	70–50

P—Pigmented, NP—Non-pigmented.

In F_2 generation a good fit of 3 : 1 in pigmented to non-pigmented ratio was obtained. This result was also confirmed by the 1 : 1 ratio in the test-cross data showing the monogenic dominance of pigmentation in the coleoptilar leaf of the seedlings of pearl millet (*Pennisetum typhoides* S and H).

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Aligarh (India), *October* 11, 1975.

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1. Gill, B. S., *Indian J. Genet.*, 1969, **29**, **468**.
2. Singh, Dharampal, Mishra, S. N., Singh, A. B. and Singh, S. P., *Ibid.*, 1967, **27**, **426**.
3. Singh, D., Lal, S. and Yadav, H. R., *J. Indian Bot. Soc.*, 1968, **47**, 388.
4. —, — and Singh, R. S., *Ibid.*, 1969, **48**, **135**.
5. Yadav, R. P., *J. Indian Bot. Soc.*, 1971, **50**, 252.

Microchemical Test for Triterpenoids in Plants

Triterpenoids occur widely in plant kingdom. Their presence is important from chemo-taxonomic as well as pharmacognostic point of view. Triterpenoids are fairly stable compounds and can be spotted in cells of free-hand sections of fresh as well as old dry specimens. Stems and roots are suitable for the purpose, but in the case of leaves and other tender parts some modifications of this method are necessary.

To study the cells containing triterpenoids, it is necessary to prepare free-hand sections, which should not be very thin and must have some intact layers of cells. The dry stems or roots may be boiled in water to make them suitable for sectioning. Using of excess of water during sectioning should be avoided and the cut sections should be trasferred to a watch-glass directly from the razor blade. It is advisable to use glass needles in the whole procedure. The selected sections are completely dehydrated by immersing in acetic anhydride for two minutes. These are then washed thoroughly in hexane by giving two changes. At this stage, the sections are divided into two batches, one for the treatment and the other as control for comparison. The first batch is treated with thyonil chloride in a dry specimen tube with cover for 2 minutes. These sections are then washed thoroughly in hexane (twice) and mounted in balsam. The control batch of sections is also mounted side by side with the treated ones for comparative study under microscope. The triterpenoid bearing cells appear olive-green, brown or violet in colour. The stain is permanent.

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Digestive Enzymes in the Pharyngeal Nephridia of *Pheretima posthuma*

The pharyngeal nephridia of the earthworm *Pheretima posthuma* are located as paired tubes in the 4th, 5th and 6th segments, among which the nephridia of the 4th and 5th segments open into the pharynx while that of the 6th segment opens into the buccal cavity. Though nephridia are primarily excretory in function[1], the pharyngeal nephridia, in view of their topographical position and in the absence of nephridial funnel are suspected to function as salivary glands.

The pharyngeal nephridia are dissected out, freed from the adherent tissue, washed in distilled water and homogenized in pyrex grinder. The enzymes are studied as per the method described earlier[2], and the hydrolized products were detected chromatographically. In these pharyngeal nephridia, amylase, melezitase, trehalase, chymosin, prolinase, prolidase, glycyl-L-leucine dipeptidase, glycylglycine dipeptidase, leucine aminopeptidase, aminotripeptidase and lipase are present but not other enzymes such as cellulase, sucrase, maltase, β-glucosidase, α-galactosidase, β-galactosidase, β-fructosidase, pepsin, trypsin and carboxypolypeptidase. Considering the presence of enzymes, the pharyngeal homogenates are able to hydrolyse a variety of substances. These experiments clearly indicate that these pharyngeal nephridia function as digestive or salivary glands.

I am indebted to Prof. R. Rakshpal, for the guidance and S.C.S.I.R., for the financial assistance.

Department of Zoology, SHRAWAN KUMAR GUPTA.
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Lucknow (India), *October* 22, 1975.

1. Bahl, K. N., *Biol. Rev.*, 1947, 22, 109.
2. Gupta, S. K., (In Press), 1975.

Free Pools of Amino Acids and Sugars in *Leptadaenia pyrotechnica* F.

Leptadaenia pyrotechnica F. (family asclepiadaceae) has some phenolic compounds and alkaloids which add to its medicinal value[1-3]. The analysis of free pools of amino acids and sugars gave the following results :

Leptadaenia pyrotechnica stem has six amino acids : lysine (8·9%), arginine (14·6%), alanine (14·7%), threonine (13·0%), methionine (9·4%), isoleucine (9·8%) and two dipeptides : *dl*-alanyl-*l*-alanine (18·5%), glycyl-*l*-alanine (11·2%). The presence of dipeptides was further confirmed by the total acid hydrolysis.

Total sugar content of the stem tissue was 8·7 mg/gm wet wt. chromatographic analysis revealed the presence of raffinose (10·8%), sucrose (33·4%), glucose (35·2%) and fructose (20·8%).

Authors are thankful to Professor A. K. Dattagupta, and Miss Neelam Ghai, for the guidance and inspiration.

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1. Subramanian, S. S., *Curr. Sci.*, 1968, 37, 373.
2. Narasimhamurthy, G., *Indian Vet. J.*, 1969, 46, 510.
3. Dantwal, A. S. and Patel, R. P., *Indian J. Pharm.*, 1958, 20, 241.

REVIEWS AND NOTICES OF BOOKS

Physics—Concepts and Models. By E. J. Wenham, G. W. Dorling, J. A. N. Snell, and S. Taylor. (Addison-Wesley Pub. Ltd., London), 1974. Pp. xi + 544. Price not given.

The book *Physics—Concepts and Models* by E. J. Wenham, G. M. Dorling, J. A. N. Snell and B. Taylor, gives an excellent reading material for students at the pre-university and undergraduate level. It brings together, the diverse topics in physics, into a mainstream of fundamental concepts which are closely akin. The reader will thus be able to link up all these ideas into a coherent network of understanding.

There are innumerable number of books on this topic; but this one scores over the others in the following: Firstly, it is a concept oriented book. The emphasis has been on the concepts and phenomena rather than on data and measurement. Perhaps this is its lacuna as well. Many simple experiments have been suggested to demonstrate a number of ideas such as vibration-forced, damped, free, etc., or collision-scattering, conservation of momentum, energy, diffraction, interference, etc. But there are a few experiments involving acurate measurements, *e.g.*, velocity of sound or light, thermal or optical properties, etc. Secondly, the difficult concepts have been discussed in a very lucid manner, *e.g.*, the distinction between inertial and gravitational mass, the different force fields, etc. Thirdly, the book has a collection of some very interesting problems which should definitely provide a challenge to the pre-university students.

The reviewer feels that some of the topics like models of liquids, thermal conductivity, radiation-black body-Stefan Boltzmann law, optical properties, acoustics-sonic as well as ultrasonic, sources, conversion and storage of energy could have been discussed in some more detail. The concept of entropy seems to have been left out. In spite of all this, the book will definitely be able to create interest in the student and also help the teacher in reorienting his lectures in a wider perspective. The general get up of the book with eyecatching pictures, is commendable.

S. V. SUBRAMANYAM.

Annual Review of Plant Physiology (Vol. 26). 1975. Pp. 551. Price: USA $ 15.00, elsewhere: $ 15.50.

In this volume of *Annual Review of Plant Physiology* three articles (1) Plasmodesmata by A. W. Robards, (2) Carbohydrates, Proteins, Cell surfaces and the Biochemistry of Pathogenesis by P. Albersheim and A. J. Anderson-Prouty, (3) Membrane Biogenesis by D. J. Morre, are published in the field of cell organization. In the Area of Bioenergetics, the Articles: Tree Photosynthesis by M. Schaedle, Thermogenic Respiration in Aroids by B. J. D. Meeuse, and Chlorophyll—Proteins: Light-harvesting and Reaction Center Components of Plants by J. B. Thornber have been published. Seven articles (1) Biochemistry of Legume Seed Proteins by A. Millerd, (2) Assimilatory Nitrate-Nitrite Reduction by E. J. Hewitt. (3) The Regulation of Carbohydrate Metabolism by J. F. Turner and D. H. Turner, (4) Plant Nucleases by C. M. Wilson, (5) Plant Sterols by C. Grunwald, (6) The Washing or Aging Phenomenon in Plant Tissues by R. F. M. Van Steveninck, (7) Phycobiliproteins and Complementary Chromatic Adaptation by L. Bogorad in the field of Metabolism are included.

The Articles, Seed Formation by L. S. Dure III; Genetic Analysis and Plant Improvement by T. B. Rice and P. S. Carbon, Apical Dorminance by I. D. J. Phillips cover the area of development and the article, Stomatal Action by K. Raschke coming under the topic of environmental physiology.

Incompatibility and the Pollen Stigma Interaction by J. Heslop-Harison and Gas Vesicles by A. E. Walsby are the special topics covered in this issue.

The Prefatory Chapter: Days of Visual Spectroscopy by Robert Hill makes an interesting reading and one can now realise the advances made in technology resulting in development of sophisticated equipment now available for research in the area of photosynthesis.

K. S. KRISHNASASTRY.

Directory of Scientific Research in Indian Universities. (University Grants Commission and Council of Scientific and Industrial Research, New Delhi), 1974. Pp. 652. Price: Rs. 35.00.

The dynamic and constantly changing nature of research activity in the Universities, Indian Institutes of Technology, Engineering and Medical Colleges and other institutions of higher learning is well reflected in this Directory of Scientific Research in Indian Universities, 1974. The joint effort of the UGC and the CSIR deserves copious thanks and appreciation from all research workers for bringing out this Directory.

The present directory is a sequel to the one published by the INSDOC in 1972 as the Current Research Projects in CSIR laboratories, in response to the need for a ready reference source of information on the organized research activities in the country. It contains some 9,910 entries which describe current research projects carried out by 11,000 research workers working in nearly 1,000 departments of the various academic institutions.

The research programmes in the Directory have broadly been arranged under seven sections according to the universal decimal classification system. The sections are, (1) Physical Sciences; (2) Bio-Sciences; (3) Engineering; (4) Agriculture; (5) Management and Transport; (6) Chemical Technology; and (7) Manufacturers, Industries, Building and Architecture.

Each section has been further divided into smaller groups and entries in the smaller groups have been arranged alphabetically by the title of the project. All the entries have been serially numbered for easy reference from the index. In addition to the assistance which is provided by the classified arrangement, the following four indexes facilitate maximum help to the seeker to approach subject of his interest in more than one way:

1. Investigator Index;
2. Key-word Index;
3. University *cum* Institution Index; and
4. Sponsor Index.

All these indexes have been arranged in alphabetical sequence with the entry number in the text.

All these indexes are excellent—necessarily so in a directory which serves as a reference source for researchers—and consequently the Directory is more useful than other types of directory which lack such indexes. These indexes contribute to the flexibility with which the user can approach the contents and the value of these as either direct or indirect routes to the desired information will be readily apparent to the user of the Directory. It is a pity that more publishers do not insist on the inclusion of extensive indexes even though the resulting documents may be more expensive, but in the long run, readers would find the extra expense well worthwhile.

Apart from its useful scientific content, the printing and get-up are excellent. It is a measure of INSDOC's achievement and a tribute to its painstaking effort that it has imposed a logical order in this otherwise chaotic output of thousands of authors. The reviewers have found the publication highly informative and they have strong reasons to believe that it will be a volume worth possessing in all the libraries of universities/institutions and research organizations. We hope that the INSDOC would continue to publish this vital service to the research community.

T. K. S. Iyengar.
G. S. R. Rao.

The General Point Processes; Applications to Structural Fatigue Bioscience and Medical Research. By V. K. Murthy. (Addison-Wesley Publishing Company, Mass, U.S.A.), 1974. Pp. xix + 604. Price: Cloth $ 22.50; Paper $ 13.50.

The book under review is the fifth one in the 'Applied Mathematics and Computations' series edited by R. Kalaba. The author's objectives are (*i*) to develop the theory of distribution of events in a specified time for a general point process. (*ii*) to bring together as many fields of application of this theory with an emphasis on the inter-disciplinary nature of this subject. This reviewer feels that the author has been quite successful.

There are twelve chapters in all. The first one introduces the notation and derives some results on the cumulants of a general point process. Chapter 2 is concerned with the properties of a proposed estimator for the variance of a point process. Some generalizations of the results of Hammersley regarding renewal processes are presented in Chapter 3. Chapters 4 and 5 deal with the moments and estimators of two-dimensional point processes. Chapter 6 applies the theory to cumulative fatigue damage in engineering structures. Inclusion of some experimental results, so widely available in the literature on cumulative fatigue, would have enhanced the utility of this chapter.

The superposition of an arbitrary number of point processes is treated in Chapter 7. The development parallels that of Cox and Smith's paper "On the superposition of renewal processes". The distribution of the number of failures, given the failure law, is presented in Chapter 8. In Chapters 9 and 10 the statistical estimation of parameters involved in some of the distributions commonly used in reliability theory is developed.

Applications to biological and medical problems are treated in Chapters 11 and 12. A critical review of the literature followed by a Markovian renewal processes model for human reproduction is presented in Chapter 11. Chapter 12 develops the solution of the renewal process integral equation by initial value methods. It also considers the theoretical basis for the stochastic analysis of electrocardiograms (E.C.G.) and brain waves (E.E.G.) data.

On the whole the book is well written and quite readable for anyone with some familiarity with probability. The strong point of the book is that it puts together many new results scattered in the literature into a neat pattern and shows how they are applicable in such widely different disciplines as engineering and medicine. However, a non-mathematician may feel that a desire to put too many things in a short space has made the presentation somewhat vague and academic. All the same research workers interested in the applications of real life problems would benefit greatly from this book. The bibliography at the end is quite valuaple but the lack of a subject index is inexcusable in a book of this kind.

R. NARAYANA IYENGAR.

Techniques and Methods of Polymer Evaluation (Vol. 4). *Polymer Molecular Weights,* Part I, Edited by Philip E. Slade, Jr. (Marcel Dekker, Inc., New York), 1975. Pp. ix + 286. Price $ 24.50.

For polymeric materials to be of practical value, their molecular weights must fall in certain fixed ranges depending on their end use requirements. Hence the molecular weights and the molecular weight distribution must be controlled. This can only be done by their measurements and a scientist working in the field of molecular weight distribution has to be conversant with the different techniques of molecular measurement and needs a ready reference book. This two-part book, entitled 'Polymer Molecular Weights' incorporates all the major techniques used to determine the molecular weights of polymers.

Part I consists of five chapters. Chapter I briefly introduces the topic of molecular weight averages with a hypothetical example. Even a beginner can very easily understand the different types of molecular weight averages after going through this chapter.

Chapter II deals with 'Membrane Osmometry'. The general design of both static and dynamic osmometers is briefly described and the various osmometers readily marketed are briefly introduced. The membranes used, their selection and the problems encountered are well presented.

The 'End group determinations' are dealt with in Chapter III. All techniques which are used to measure polymer end groups of synthetic polymers are presented with the necessary experimental details and critically reviewed.

'Absolute Colligative Property Methods' form the topic of Chapter IV. Methods such as ebulliometry, cryoscopy, differential vapour pressure, and vapour pressure osmometry used for the determination of the number average molecular weights of synthetic polymers are described in detail.

'Light Scattering from Solutions of Macromolecules' constitutes the subject-matter of Chapter V. This chapter deals at length with the theory, the light-scattering methods and the light-scattering instrumentation. A brief mention about the recently burgeoning studies of small shifts in wavelength in scattering, which are of interest in connection with molecular dynamics is also made.

Each chapter is complete by itself and deals at length with the pertinent theory, instrument design, a critical survey of the commercially available instruments, calibration procedures, experimental precautions to be taken, data analysis and references. In the data analysis, if typical numerical examples and the graphical procedures involved were also included, the usefulness of the book would have increased considerably even to technicians not concerned with the theory. Although the book is published in 1975 the references listed in most of the chapters are up to 1971 and very few references of 1972 have been included.

After the publication of 'Techniques of Polymer Characterisation' edited by P. W. Allen, in 1959; and 'Newer Methods of Polymer Characterisation' in *Polymer Reviews,* Vol. 6, edited by B. Ke, in 1964, to the author's best knowledge there is no other book dealing with the techniques and methods of polymer evaluation excepting a few review articles on some representative topic in some journals and series publications. In the last two decades tremendous progress has been achieved in improving the existing experimental techniques and data analysis. Hence this book forms a welcome addition and makes it the most convenient source of information on molecular weight measurement for polymer chemists and physicists, and scientists in the plastics and synthetic fiber industries.

V. KALPAGAM.

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COBALT (II) COMPLEXES WITH TRIDENTATE LIGANDS CONTAINING -0, 0, 0-SEQUENCES

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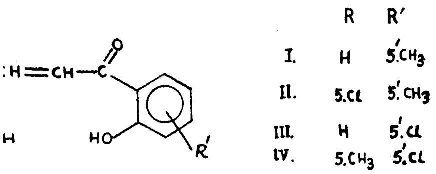
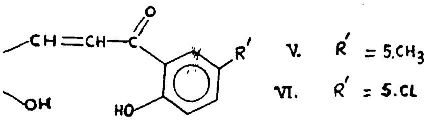
INTRODUCTION

t for tridentate ligands containing a ting sequence which is different from O, N, O; O, N, S; and N, N, N we have attempted a new series of aining 0, 0, 0 sequence, on which very little 2, 2′-dihydroxy-substituted-chalkones con- class of ligands. In this communica- ish to report synthesis and spectral f a few cobalt (II) complexes with the below.





EXPERIMENTAL

hemicals used for preparing the chalkones t acetate were of reagent grade. Sub- ehydes were prepared according to Duff's Chalkones were prepared by the method the literature[4].

plexes were prepared by treating disodium s with cobalt (II) acetate in 1 : 1 molar in aqueous-alcohol medium. The com- arated out were filtered, washed with dried at 110° C. The dried complexes dered and extracted with cyclohexane in thimble in order to free them from ligand. All the complexes were dried at 110° C. lexes were analysed for their cobalt con- only three complexes were got analysed and hydrogen contents. The results are Table I.

RESULTS AND DISCUSSION

sults of the elemental analysis (Table I) t the cobalt (II) complexes have 1 : 1 try.

TABLE I

Elemental analysis of Cobalt (II) complexes with 2, 2′-dihydroxy chalkones

Lig. No.	Empirical Formula	% Co	% C	% H
I.	$(C_{16}H_{12}O_3)Co \cdot H_2O$	18·15 (17·94)	57·85 (58·35)	3·57 (4·25)
II.	$(C_{16}H_{11}Cl \cdot O_3)Co \cdot H_2O$	15·91 (16·24)	..	..
III.	$(C_{15}H_9Cl \cdot O_3)Co \cdot H_2O$	17·00 (16·89)	50·85 (51·51)	3·24 (3·15)
IV.	$(C_{16}H_{11}Cl \cdot O_3)Co \cdot H_2O$	16·35 (16·24)	..	..
V.	$(C_{20}H_{14}O_3)Co \cdot H_2O$	15·33 (15·55)	62·05 (53·32)	4·14 (4·22)
VI.	$(C_{19}H_{11}Cl \cdot O_3)Co \cdot H_2O$	14·45 (14·75)	..	..

The values in the parenthesis are the calculated values.

Magnetic data.—The magnetic susceptibilities of the complexes have been measured on a Gouy balance at room temperature. The μ eff. values fall in the range of 4·5 to 5·0 B.M. indicating that cobalt (II) has tetrahedral configuration in these complexes.

Electronic spectra.—The electronic spectra of the complexes have been measured in DMF at room temperature on a Beckman DB-2 recording spectrophotometer.

The cobalt (II) complexes show multi component band in the region 450–700 nm, due to the tetrahedral configuration and the band is assigned as $3\,A_2 \rightarrow 4\,T_1$ (P) transition.

Infrared spectra.—The infrared spectra of the ligands and the complexes in Nujol mull have been recorded on a Beckman IR-18.

In all the ligands spectra, we observe two bands, one in the region 3300–3200 cm^{-1} (medium intensity) and the other in the region 2650–2600 cm^{-1} (weak and broad) and these have been attributed respectively to the inter- and intra-molecular hydrogen bonded –OH groups in view of the previous assignments[6]. In the complexes, both the bands disappear showing that both the hydroxy groups are involved in the bond formation. We observed a sharp broad

TABLE II

Important infrared frequencies (in cm^{-1}) of chalkones and their cobalt (II) complexes

Sl. No.	2, 2′-dihydroxy 5′-Me-chalkone L	C	2, 2′-dihydroxy 5-Cl-5′-Me-chalkone L	C	2, 2′-dihydroxy 5′-Cl-chalkone L	C	Assignments
1.	3200 m.	..	3200 br.m.	..	3300 m.	..	Intermolecular H–bonded OH
2.	2650 br.w.	..	2650 br.w.	..	2675 br.w.	..	Intramolecular H–bonded OH
3.	1640 s	1626 s.	1648. s.	1626 s.	1650 s.	1639 s.	C=O stretch
4.	1595 m.	1600 m.	1600. s.	1613 m.	1600 m.	1613 br.w.	Ethylenic C=C stretch
5.	1570 s 1495 m 1480 s	1550 m. 1527 br. m. 1478 m.	1558. s. .. 1470. s.	1565 m. 1534 m. 1470 s.	1550 s. .. 1470 s.	1575 s. .. 1471 s.	Aromatic C=C stretch
6.	1265 s	1336 m.	1280. s.	1316 s.	1280 m.	1307 m.	Phenolie C–O stretch
7.	..	3356 br.m.	..	3450 br. m.	..	3450 br. m.	Coordinated water

TABLE II (*Contd.*)

Sl. No.	2, 2′-dihydroxy 5-Me-5′-Cl-Chalkone L	C	2, 2′-dihydroxy 5, 6-benzo-5′-Me-Chalkone L	C	2, 2′-dihydroxy 5, 6-benzo-5, 6-benzo-5′-Cl L	C
1.	3250 m.	..	3250 m.	..	3250 m.	..
2.	2675 br.w.	..	2650 br.w.	..	2650 br.w.	..
3.	1648 s.	1639 s.	1650 s.	1634 s.	1648 s.	1626 s.
4.	1590 s.	1613 m.	1600 m.	1613 br. m.	1600 m.	..
5.	1575 m. 1525 w. 1470 s.	1587 w. 1562 m. 1480 s.	.. 1625 m. 1470 s.	1575 s. 1527 w. 1470 s.	1575 m. 1525 w. 1470 s.	.. 1538 m. 1515 s. 1470 s.
6.	1280 s.	1316 s.	1290 s.	1307 s.	1285 s.	1333 s.
7.	..	3480 br.m.	..	3480 br.m.	..	3448 br.m.

L=Ligand; C=Complexes; s=strong; m=medium; w=weak; br=broad.

band in the region 3480–3350 cm^{-1} which is indicative of coordinated water molecule. No loss in the weight of the complex has been observed even after heating for 2 to 3 hours at 120° C. Removal of the water molecule at elevated temperature results in the decomposition of the complex. These results suggest that the water molecule present in the complex is not loosely bound.

An intense band observed for ligands in the region 1650–1639 cm^{-1} attributable to the C=O stretching vibration[7] is observed in the region 1639–1626 cm^{-1}, for the complexes. This shift towards the lower frequency may be due to the coordination of the C=O group to the cobalt (II) through oxygen which ultimately results in decreasing the bond order of carbon to oxygen link.

The medium to high intensity bands appearing in the region, 1580–1460 cm^{-1} for both ligands and the complexes, are assigned to the aromatic C=C stretching vibrations. The phenolic C—O vibration in the region 1280–1265 cm^{-1} of the ligands is seen in the region 1336–1307 cm^{-1} in the complexes supporting the conclusion that OH groups are involved in the bond formation.

Thus the analytical, magnetic, electronic and infrared data suggest that cobalt (II) has a coordination number of four with the tetrahedral configuration.

The construction of the model for the complexes indicates that the monomeric structure involves much steric hindrance as the ligand molecule has to attach itself to the one and the same metal ion through all its coordinating sites. The low solubility of the complexes in the common organic solvents may be due to an extended polymeric chain structure.

1. a. Holm, R. H., Everette (Jr.), G. W. and Chakravorty, A., *Progr. Inorg. Chem.*, 1966, 7, 83.
   b. Muto, Y., *Bull. Chem. Soc., Jap.*, 1958, 31, 1017.
   c. Sacconi, L., *Coord., Chem. Rev.*, 1966, 1, 126 and 192.
   d. —, Speroni, G. P. and Morasi, R., *Inorg. Chem.*, 1968, 7, 1521.
   e. Lindoy, L. F. and Livingston, S. E., *Ibid.*, 1968, 7, 1149.
   f. Lee, C. C., Syamal, A. and Theriot, L. J., *Ibid.*, 1971, 10, 1669.
2. a. Evard, F., *Compt. Rend.*, 1933, 196, 2007.
   b. Tsumaki, T., Murase, I. and Nishimura, T., *J. Chem. Soc., Jap.*, 1951, 72, 368.
   c. Tolmachev, V. N., Boberov, C. F. and Luvrushin, V. F., *Dopov, Akad, Nauk, Ukr. R.S.R.*, 1966, p. 1454; *C.A.*, 1967, 66, 60470 *a*.
   d. —, Yushko, V. K. and Luvrushin, V. F., *Zh. Obshch. Khim.*, 1970, 40, 1518; *C.A.*, 1971, 74, 63812 *f*.
3. Duff, J. C., *J. Chem. Soc.*, 1941, p. 541.
4. Dhar, D. N., *J. Indian Chem. Soc.*, 1960, 37, 363.
5. Cotton, F. A. and Wilkinson, G., *Advanced Inorganic Chemistry*, Wiley Eastern, New Delhi, 1969.
6. a. Rao, C. N. R., *Chemical Applications of Infrared Spectroscopy*, Academic Press, London, 1958.
   b. Ramaswamy, K. K., Jose, C. I. and Sen, D. N., *Indian J. Chem.*, 1967, 5, 156.
7. Dhar, D. N. and Gupta, V. P., *Ibid.*, 1971, 9, 818.

POLAROGRAPHIC STUDY OF BIOLOGICALLY IMPORTANT COMPLEXES : Cd (II) AND Zn (II) COMPLEXES WITH CREATININE

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ABSTRACT

Complexation of creatinine with cadmium and zinc has been studied in aqueous medium at 25 ± 0·5° C. Creatinine formed three complexes with Cd (II) with their respective stability constants : $\log \beta_1 = 1{\cdot}48 \pm 0{\cdot}025$, $\log \beta_2 = 1{\cdot}60 \pm 0{\cdot}02$ and $\log \beta_3 = 2{\cdot}07 \pm 0{\cdot}035$. Two complex species were observed with Zn (II) with their respective stability constants : $\log \beta_1 = 0{\cdot}60$ and $\log \beta_2 = 2{\cdot}14$ (approx.). Zn(II)-creatinine system was examined by the method of Ringbom and Eriksson.

INTRODUCTION

CREATININE, an anhydride of creatine, represents a waste product of creatine metabolism in the body. Studies on the binding of metal ions with creatinine are of topical interest since they may be used as models for the study of metal-creatine interactions.

Literature survey revealed that there is no reported study on the polarographic determination of the nature and stability of metal complexes of creatinine except that of Pleticha's work on creatinine complexes with the iron group metals[1]. This paper deals with the evaluation of the stability constants of Cd(II)-creatinine and Zn(II)-creatinine complexes.

EXPERIMENTAL

All chemicals used were of reagent grade. Sodium nitrate was used to maintain ionic strength constant at 1·0 M. All solutions were prepared in doubly distilled water. The dropping mercury electrode had the following characteristics:

$$t = 3{\cdot}21 \text{ sec in } 0{\cdot}1 \text{ M. } NaNO_3 \text{ (open circuit)}$$
$$\text{and}$$
$$m = 2{\cdot}18 \text{ mg. sec.}^{-1}.$$

Measurements were made on Cambridge automatic recording polarograph. The half-wave potentials were obtained from the log-plots. Deoxygenation of the solutions to be polarographed was achieved by nitrogen gas. A saturated calomel electrode (SCE) was used as a reference electrode.

RESULTS AND DISCUSSION

Cd (II)-Creatinine System

A series of solutions of 1m M Cd(II) with various amounts of creatinine (0·08 to 0·40 M) and the requisite

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COBALT (II) COMPLEXES WITH TRIDENTATE LIGANDS CONTAINING -0, 0, 0-SEQUENCES

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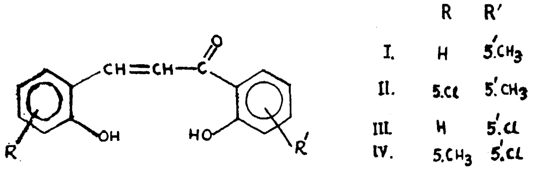
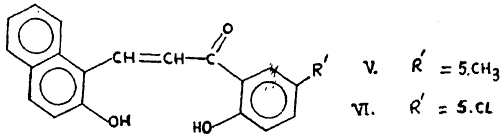
INTRODUCTION

IN a quest for tridentate ligands containing a coordinating sequence which is different from the known O, N, O; O, N, S; and N, N, N sequences[1], we have attempted a new series of ligands containing 0, 0, 0 sequence, on which very little is known[2]. 2, 2′-dihydroxy-substituted-chalkones constitute this class of ligands. In this communication we wish to report synthesis and spectral properties of a few cobalt (II) complexes with the bases shown below.





EXPERIMENTAL

All the chemicals used for preparing the chalkones and cobalt acetate were of reagent grade. Substituted aldehydes were prepared according to Duff's method[3]. Chalkones were prepared by the method reported in the literature[4].

The complexes were prepared by treating disodium chalkonates with cobalt (II) acetate in 1 : 1 molar proportion in aqueous-alcohol medium. The complexes separated out were filtered, washed with water and dried at 110° C. The dried complexes were powdered and extracted with cyclohexane in a soxhlet thimble in order to free them from ligand impurities. All the complexes were dried at 110° C. The complexes were analysed for their cobalt content and only three complexes were got analysed for carbon and hydrogen contents. The results are shown in Table I.

RESULTS AND DISCUSSION

The results of the elemental analysis (Table I) show that the cobalt (II) complexes have 1 : 1 stoichiometry.

TABLE I

Elemental analysis of Cobalt (II) complexes with 2, 2′-dihydroxy chalkones

Lig. No.	Empirical Formula	% Co	% C	% H
I.	$(C_{16}H_{12}O_3)Co \cdot H_2O$	18·15 (17·94)	57·85 (58·35)	3·57 (4·25)
II.	$(C_{16}H_{11}Cl \cdot O_3)Co \cdot H_2O$	15·91 (16·24)	..	..
III.	$(C_{15}H_9Cl \cdot O_3)Co \cdot H_2O$	17·00 (16·89)	50·85 (51·51)	3·24 (3·15)
IV.	$(C_{16}H_{11}Cl \cdot O_3)Co \cdot H_2O$	16·35 (16·24)	..	..
V.	$(C_{20}H_{14}O_3)Co \cdot H_2O$	15·33 (15·55)	62·05 (53·32)	4·14 (4·22)
VI.	$(C_{19}H_{11}Cl \cdot O_3)Co \cdot H_2O$	14·45 (14·75)	..	..

The values in the parenthesis are the calculated values.

Magnetic data.—The magnetic susceptibilities of the complexes have been measured on a Gouy balance at room temperature. The μ eff. values fall in the range of 4·5 to 5·0 B.M. indicating that cobalt (II) has tetrahedral configuration in these complexes.

Electronic spectra.—The electronic spectra of the complexes have been measured in DMF at room temperature on a Beckman DB-2 recording spectrophotometer.

The cobalt (II) complexes show multi component band in the region 450–700 nm, due to the tetrahedral configuration and the band is assigned as $3\,A_2 \rightarrow 4\,T_1$ (P) transition.

Infrared spectra.—The infrared spectra of the ligands and the complexes in Nujol mull have been recorded on a Beckman IR-18.

In all the ligands spectra, we observe two bands, one in the region 3300–3200 cm^{-1} (medium intensity) and the other in the region 2650–2600 cm^{-1} (weak and broad) and these have been attributed respectively to the inter- and intra-molecular hydrogen bonded –OH groups in view of the previous assignments[6]. In the complexes, both the bands disappear showing that both the hydroxy groups are involved in the bond formation. We observed a sharp broad amount of supporting electrolyte were polarographed at 25 ± 0·5° C. Maximum supressor was not needed. Half-wave potentials and reversibility of the current-voltage curves were obtained from the plots of

$-E_{d.e.}$ *vs*, log $i/(id - i)$.

A plot of $-E_{\frac{1}{2}}$ *vs.* log C_L revealed the existence of three complexes. DeFord and Hume's method[2] was, therefore, applied to evaluate the stability constants (Fig. 1) of these complexes which are given below:

$\log \beta_1 = 1{\cdot}48 \pm 0{\cdot}025$, $\log \beta_2 = 1{\cdot}60 \pm 0{\cdot}02$ and $\log \beta_3 = 2{\cdot}70 \pm 0{\cdot}035$.

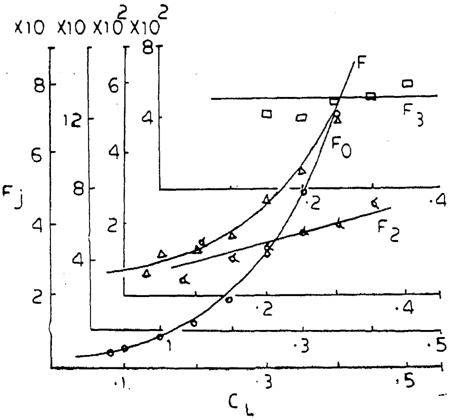


FIG. 1. Plots of F_j *vs.* C_L.

Zn(II)–Creatinine System:

The system was investigated by "indicator ion" method[3]. Cd(II), reducing at a potential more positive than that of Zn(II), was chosen as the indicator ion. As the half-wave potential for Cd(II) in presence of creatinine and at ionic strength of 1·0 M was required for comparison purposes under the present experimental conditions, to determine the shift in the presence of Zn(II), the half-wave potentials of Cd(II) in presence of different concentrations of creatinine were taken from the data of Cd(II)–Creatinine system.

These half-wave potentials are represented graphically in Fig. 2 (*a*). The reversibility of the electrode reaction and the half-wave potentials were determined from the log-plots. The concentration of free ligand was taken to be the same as the total creatinine concentration, as Cd(II) formed rather weak complexes.

Polarograms of solutions containing 1m M Cd (II) and 0·14 M zinc were taken at different concentrations of creatinine. When zinc was added to a solution containing cadmium and creatinine, the half-wave potential of Cd (II) shifted to more positive potentials due to the decrease in free ligand concentrations as a result of the formation of zinc complexes. This is graphically represented in Fig. 2 (*b*).

The half-wave potentials were read from the smooth curve to obviate any experimental error in the individual measurements and are set out in Table I.

TABLE I

Free creatinine and $\bar{n}$ *values for Zn (II)-creatinine system*

C_L M	$-E_{\frac{1}{2}}$ vs. S.C.E.	[L] M	$\bar{n}$ exptl.
	Zn(II) = 0·14 M		
·025	·601	·010	0·11
·040	·603	·020	0·13
·050	·603	·025	0·18
·070	·605	·030	0·28
·100	·607	·040	0·43
·125	·609	·050	0·53
·150	·610	·055	0·68
·175	·612	·070	0·75
·200	·613	·075	0·89
·225	·614	·080	1·04
·250	·615	·085	1·18
·275	·616	·090	1·32
·300	·616	·090	1·50
	Zn(II) = 0·22 M		
·05	·602	·020	0·13
·09	·604	·030	0·27
·11	·606	·035	0·34
·15	·608	·040	0·50
·17	·608	·045	0·57
·19	·609	·050	0·64
·21	·610	·055	0·70
·25	·611	·070	0·82
·33	·612	·074	1·16

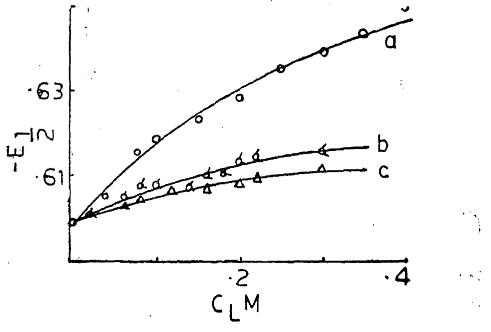


FIG. 2. Plot of $-E_{\frac{1}{2}}$ *vs.* C_L for Zn(II)-creatinine system; *a*–Cd(II), *b*–Cd(II) + 0·14 M Zn(II), *c*–0·22 M Zn(II).

The concentration of free ligand was calculated from these half-wave potentials using a plot of $E_{\frac{1}{2}}$ *vs.* concentration of creatinine for Cd (II) by following the usual procedure (Fig. 2). These values are presented in Table I. The metal-ligand formation number $\bar{n}$ is defined as:

$$\left(\frac{C_L - [L]}{C_{Zn}}\right)$$

Where C_L is initial concentration of creatinine, [L] is the free ligand concentration and C_{Zn} is the amount of zinc added. The values of $\bar{n}$ thus calcualated are plotted against [L].

TABLE II
Values for Rossotti Plot

[L] M	$\bar{n}$ exptl.	$\bar{n}$ (Calcd.)	$\frac{\bar{n}}{(1-\bar{n})[L]}$	$\frac{(2-\bar{n})}{(1-\bar{n})}[L]$
0·030	·26	·29	11·71	·070
.034	·33	·35	14·49	·085
.036	·37	.38	16·31	·093
·038	·40	·41	17·54	·101
·040	·43	·43	18·86	·110
·048	·54	·55	24·46	·152
·054	·62	·63	30·21	·196
·060	·70	·71	38·89	·260
·066	·74	·79	43·12	·320

These results were further confirmed by using a different concentration of zinc. Polarograms of solutions containing 1 mM Cd(II) and 0·22 M Zn(II) were taken at different concentrations of creatinine. The results are given in Table I and are represented graphically at *c* in Fig. 2. $\bar{n}$ values calculated in the manner described above are also plotted with the previous set of $\bar{n}$ values. There is an excellent agreement between the values obtained for two concentrations of zinc. The values of $\bar{n}$ obtained from the smooth curve of $\bar{n}$ *vs.* [L] are solved for the overall stability constants by the method of Rossotti and Rossotti[4] (Table II). The existence of two complexes was revealed from the Rossotti plot. The stability constants are found to be: $\log \beta_1 = 0{\cdot}60$ and $\log \beta_2 = 2{\cdot}14$ (Approx.).

By using these β values $\bar{n}$ values were calculated from Rossotti's equation. The calculated $\bar{n}$ values are presented along with experimental values in Table II. The agreement between two sets of $\bar{n}$ values is fairly satisfactory.

ACKNOWLEDGEMENT

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1. Pleticha, R., *Chem. Listy.*, 1951, **45**, 185.
2. DeFord, D. D. and Hume, D. N., *J. Am. Chem. Soc.*, 1951, **73**, 5321.
3. Ringbom, A. and Eriksson, L., *Acta Chem. Scand.*, 1953, **7**, 1105.
4. Rossotti and Rossotti, *Ibid.*, 1955, **9**, 1166.

THE KIRLIAN PHANTOM LEAF

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ABSTRACT

A leaf from which a small portion was excised was sandwiched between a pair of X-ray films and subject to AC pulses of about 10 KV and 20 KHz. The resultant Kirlian photographs contained, besides the expected images of the intact portion of the leaf, the 'Kirlian Phantom'—a luminous cap roughly of the same size and shape as the cut away portion of the leaf and occupying roughly the same area.

WHAT IS KIRLIAN PHOTOGRAPHY ?

DURING the past three or four years there has been a considerable interest in an imaging technique known as Kirlian Photography—named after the Russian inventor Semyonof Kirlian[1-5]. It is also referred to as Corona Discharge Photography and High Voltage Photography. In this technique, the object to be imaged is placed in a high voltage, high frequency electric field. The image is record either directly on a photographic film placed in the electric field or through a camera if transparent electrodes are used[2]. The only source of illumination is provided by the electric fields. The Kirlian image resembles the conventional image—as regards the general shape and contours of the object, but, characteristically, is made up of a large number of luminous points or streaks referred to in popular literature as 'balls of light' or 'aura'. The systematic studies of Professor Tiller and his associates at Stanford University attribute these luminous points to the streamers or coronas from the object points[2].

The recent interest in Kirlian photography has been triggered by the reports that the Russians have discovered some very strange—almost incredible—phenomena with the aid of this technique and that they have tremendous possibilities. The most talked about Kirlian phenomenon is the 'Phantom Leaf Effect'—the subject of this investigation. In the wake of the initial excitement, the systematic studies in U.S.A., notably by Tiller's group, seem to indicate that some of these popular claims may be rather exaggerated[2,4,6].

What kind of information can the Kirlian image of the object studied ? Our systematic studies using objects of varying structures and experimental configurations are aimed at answering this question empirically without getting into the complicated mechanisms of the streamer formation. This paper will, however, be confined to our work on the interesting Phantom Leaf Phenomenon.

EXPERIMENTAL TECHNIQUES

Figure 1 sketches schematically the Kirlian arrangement used by us at the IIT, Madras. A Tesla Coil built by the High Voltage Section of the IIT Electrical Engineering Department provided the AC field—about 10 KV, 20 KHz in the form of pulses of roughly 1 m sec duration, repeated at intervals of about 1 sec. The electrodes *E* were generally stainless steel plates with polished plane surfaces. A pair of dielectric sheets *D* (usually, exposed X-ray films) were used to prevent the shorting of the electrodes and also to control the field between them. The object *O* (a leaf in this investigation) and one or more recording films *F* were tightly wrapped in a black paper casette *C*: Normally, all the above surfaces were in contact. It was possible, however, to adjust the separation between one object surface and the photographic film closest to it by interesting spacers. The spacers were thin strips of exposed photographic films about 100 microns thick. Two Orwo X-ray films with emulsions on both sides were used for this investigation. The leaves used for our Kirlian experiments were young leaves freshly plucked from trees or shrubs near the laboratory. The species was considered unimportant—at least at this stage. The leaves were ensured to be reasonably flat, *i.e.*, free from curling. The best pictures were obtained with leaves about ·1 to ·2 mm thick. The maximum dimensions of the leaves were in the range—1·5 to 2·5″. The lower limit was to ensure enough area for ease of observation. The upper limit was to ensure clear spaces in the photographic film of about 1″ around the leaf boundary.

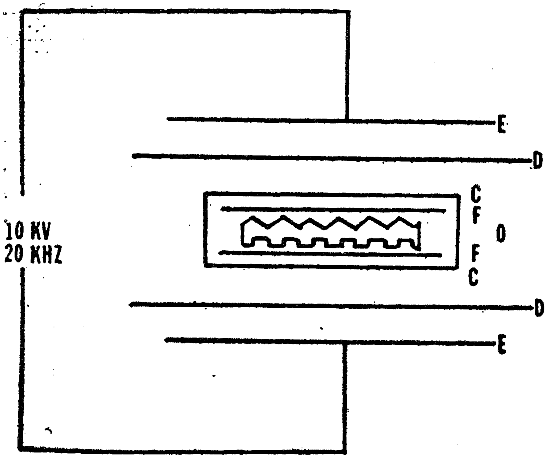

FIG. 1. Schematic representation of the Kirlian set up in the IIT, Madras. *E*, Electrodes with polished plane surfaces (Steel plate about 6 inches in diameter); *D*, Exposed X-ray films; *C*, Light proof black paper casette; *F*, Photographic films; *O*, object (leaf in this instance). The electrodes are connected to a Tesla coil. The surfaces *E*, *D*, *C*, *F* and *O* are all in contact unless separated by spacers of known thickness.

A small portion of the leaf was excised within minutes after plucking it from the plant. The excised portion was stored away and the cut leaf was sandwiched between a pair of Orwo X-ray films and wrapped in a black paper casette and placed in the electric field. After two pulses, the films were developed. A contact shadowgraph of the cut leaf was made by placing it on a fresh photographic film and shining it with light from a tungsten lamp. A similar contact shadowgraph was also made of the excised portion of the leaf.

RESULTS

The image A of Fig. 2 is the contact shadowgraph of a cut leaf used in one of our Kirlian experiments. The images B and C are the Kirlian images of the same cut leaf—one on either side. In B and C, the profiles of the leaf and the veins can be recognized. The 'V' cut can also be easily identified. The pictures are positive prints—*i.e.*, the luminous portions appear white and the shadows dark. If the Kirlian images and the shadowgraph are compared carefully, a luminous region enveloping the leaf boundary can be recognized. It can be seen as an intensely luminous cap above the 'V' cut—in the area where the cut portion of the leaf would have been located. This apparent 'filling up' of the missing portion of the cut leaf in the Kirlian image is referred to as the 'Phantom Leaf Effect'.

We have described one of our most successful experiments in imaging the Kirlian phantom leaf. Not all our experiments were so successful, however. We have not yet been able to identify the parameters which will help reproduce the phenomenon at will. For want of space we will not attempt a detailed discussion of all our results.

DISCUSSION

So far as we know, there is no published account of the phantom leaf phenomenon. The only information we have is derived from the references already cited[1,3,5,6] and from personal communications from Tiller and from Thelma Moss. None of these references, however, has reproduced any

Phantom Leaf Photograph. According to these reports[5], the phantom leaf effect is extremely elusive and has so far never been reproduced at will. Occasionally a 'near miss' is obtained. Only rarely a convincing Phantom Leaf effect is obtained. According to Tiller[6], most of these 'hits' and 'near misses' could be explained away as artefacts due to poor experimental procedures. He is satisfied with the experimental procedures only in two cases—a videotape result of Thelma Moss's group at UCLA and a sheet film result of Wagner (private communications). In both cases, the Phantom Leaf was seen : there was a light pattern in the cut away portion but it was of a distinetly different quality than in the remaining leaf region. We had no chance to see either of these two results as they have not been published. Our own Kirlian experiments were carried out with great care and all our experimental parameters have been specified in this paper. In particular, we must emphasize that the whole leaf was never brought close to the films, the casette, the electrodes or the dielectric sheets. The leaf was placed in the film sandwich only after a portion of it was already excised.

To explain the phantom leaf effect the Russians are said to have postulated the existence of an 'energy body' or 'bioplasma' associated with living objects[1,5]. It is as if the Kirlian technique brings to light the 'energy body' which would otherwise not be recognized by our sense apparatus. In other words, the 'energy body' corresponding to the cut away portion of the leaf persists for a while after the excision ; it is not perceived by our normal senses but is revealed in the Kirlian image. The question, however, is—is there any compelling necessity at the moment to postulate concepts like 'bioplasma' that do not fit in with the accepted ideas of contemporary science ? Have we exhausted alternate, simpler explanations based on known principles ?

According to Tiller[6], the luminous cap may be due to discharges from the ionized gas emissions from the solid leaf. These emissions may (*a*) be caused by vaporization by the arc channels and (*b*) by a complex process, be confined to the area of the excised portion. We are looking for an explanation in terms of the shape of the air gap around the cut away portion without invoking gaseous emissions from the solid leaf. However, as of now, these suggestions do not account for the shape and size of the luminous cap and as such, do not amount to a concrete explanation. We do hope that further work in our laboratory and elsewhere will soon lead to a better understanding of the nature and significance of this interesting phenomenon.

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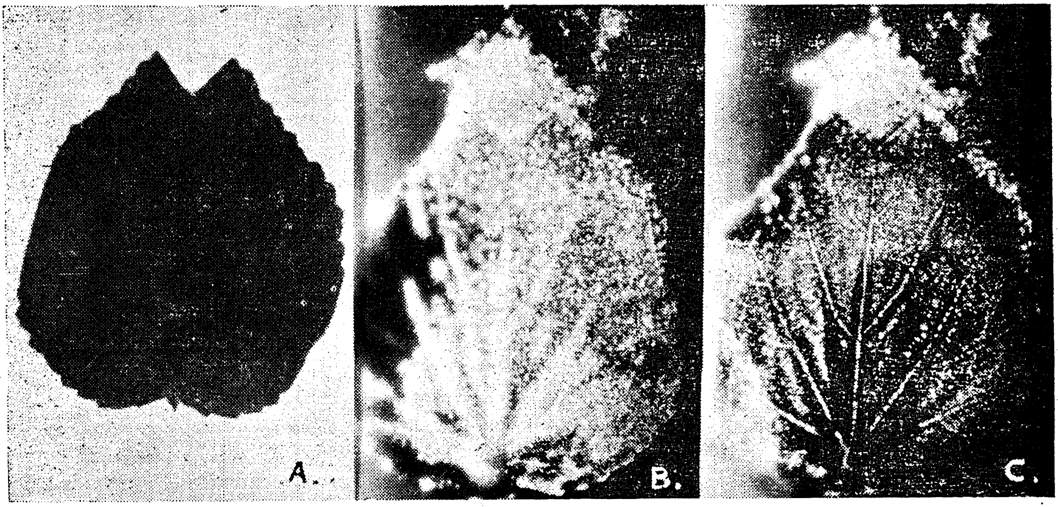

FIG. 2. Kirlian Photographs of a leaf. A, Contact shadowgraph of the cut leaf ; B, Kirlian image of the front side of the cut leaf ; C, Kirlian image of the rear side of the cut leaf. The pictures are positive prints : luminous portions appear white and shadows appear dark. The intensely luminous portion above the 'V' cut in B and C is the 'Kirlian Phantom Leaf'.

Dr. C. Narayana Reddy of the IIT Electrical Engineering Department for most generously making available the high voltage source and the ancillary facilities.

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1. Ostrander, S. and Schroeder, L., *Psychic Discoveries Behind the Iron Curtain*, Prentice Hall, New York, 1970.
2. Boyers, D. G. and Tiller, W. A., *J. Appl. Phys.*, 1973, 44, 3102.
3. Aaronson, S., *The Sciences*, 1974, p. 15.
4. Tiller, W. A., *The New Scientist*, 1974, p, 160.
5. Thelma Moss, *Phantom Leaf Effect as Revealed through Kirlian Photography*, Report from UCLA Neuropsychiatric Institute, 1974.
6. Tiller, W. A., *Psychic.*, 1975, 6, 62.

NEURAL REGULATION OF PROTEINS IN THE SKELETAL MUSCLES OF FROG, *RANA HEXADACTYLA* (LESSON)

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ABSTRACT

Electrophoretograms of normal and denervated gastrocnemius and peronius muscles of frog showed the existence of 6 bands, which are discernable into four relatively slow moving and two relatively fast moving types. Following neurectomy, the changes in the protein fractions were conspicuous in both the muscles. The atrophy effects were more prominent in the peronius than in the gastrocnemius muscle. The probable significance of the protein characters on neurectomy have been discussed.

THE effect of innervation on the biochemical, metabolic and physiological properties of the muscle has received considerable attention[1-7]. However there are only few reports on the changes in the electrophoretic pattern of cell proteins following peripheral nerve section. The present study of the denervation induced changes in the protein fraction of the muscle was undertaken to have a better understanding of the range of control exerted by the nervous system on the innervating tissues, since this will throw light on the mechanism of the neural regulation of the biochemical attributes of skeletal muscle[8,9].

Two muscles were chosen for the present study, namely, (1) the gastrocnemius, which is a mixed muscle with high proportion of slow fibres and (2) the peronius, which is essentially a fast muscle, both being innervated by the common sciatic nerve[10].

MATERIALS AND METHODS

Rana hexadactyla were subjected to unilateral denervation by severing about one centimeter of the sciatic nerve from its origin on one side of the leg, while the contralateral muscle was considered as the control. They were fed *'ad lib'* with earthworms and water was changed regularly. One month post-operatively, they were sacrificed by pithing, the gastrocnemius and the peronius muscles were isolated with least injury and washed in amphibian Ringers' medium[11]. The cell proteins were extracted as previously described[12].

Polyacrylamide disc electrophoresis was conducted by the method of Davis and Orstein[13]. 0·1 ml protein extract was directly applied, followed by a small quantity of 40% sucrose solution. A direct current of 1·5 m Amps per tube was applied for 60 minutes at 4° C in 0·05 M tris-glycine buffer at pH 8·9. After electrophoretic run, the gels were removed and stained in 1% amido black in 7% acetic acid. The excess stain was removed by repeated washings with 7% acetic acid until the non-protein part of the gel became transparent.

RESULTS AND DISCUSSION

The electrophoretic mobility patterns of cell proteins of the gastrocnemius muscle revealed the existence of 6 bands. 3 of the bands are relatively slow moving type (*a*, *b* and *c*), one is intermediary (*d*) and the remaining two are fast moving types (Fig. 1). The pattern remains similar in the denervated muscle suggesting that the major classification of the cell proteins remains unchanged. However some of the protein bands indicate physical changes involving electromobility and the quantitative aspects. The bands *'d'*, *'e'* and *'f'* in the denervated peronius showed a tendency of increased electromobility while in the gastrocnemius similar changes could not be evinced on denervation. However on the quantitative point of view, as visualized in terms of the density of the band it is likely that bands *'d'* *'e'* and *'f'* could be increasing on denervation in both the muscles. Thus the increase in fast moving fractions in both types of muscles on denervation atrophy compared to the respective controls seemed to be an essential consequence of neurectomy, and similar changes have been reported previously[9, 12]. The apparently negligible change in the protein level of the *'a'*, *'b'* and *'c'* bands may indicate the lack of neurotrophic control on them and as suggested by Guth and Watson[9] they may be under the control of intrinsic factors present in the muscle[9].

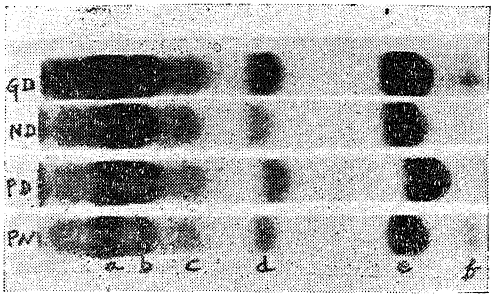

FIG. 1. Polyacrylamide disc electrophoretograms of soluble protein fractions of normal and denervated skeletal muscles, *viz.*, Gastrocnemius denervated (G.D.), Gastrocnemius normal (G.N.), Peronius denervated (P.D.), and Peronius normal (P.N.). The markings *a, b, c, d, e* and *f*, indicate protein fractions.

By comparison with the electromobility pattern of the frog serum it is likely to consider bands *'e'* and *'f'* to belong to the albumin and pre-albumin-types while *'a'*, *'b'*, *'c'* and *'d'* belong to that of globulin-type protein. The band *'d'* is comparable to the globulin-type of protein. The increase in the globulin-type of cell proteins especially that of *'d'* band may correspond to the increased proteolytic enzyme content which is globulin in nature[14]. Similar positive modulation of proteolytic enzymes by globulin-type proteins was reported in the amphibian gastrocnemius muscle[15] and the elevated protease activity in the denervated muscle could be under similar situation.

The elevated albumin and prealbumin-types as revealed by the density of *'d'* and *'f'* bands may probably contribute to the divalent ion complex and the increased Ca^{++} content reported in the denervated muscle is in agreement[16]. Even though the sciatic nerve innervates both the peronius (fast) and gastrocnemius (mixed) muscles, the denervation-induced response appear to be different in the fast muscle as compared to the mixed muscle.

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1. Gutmann, E., In : *Methods of Neurochemistry*, Ed., Fried, R., M., Dekker, Inc., New York, 1973, 15, 189.
2. Guth, L., *Physiol. Rev.*, 1968, 48, 645.
3. Gutmann, E., *The Denervated Muscle*, Czeckoslovak Academy of Sciences, Prague, 1962.
4. —, *Scientia*, 1969, 61, 1.
5. —, *Nova acta leopoldina.*, 1973, 211, 193.
6. Guth, L., In : *Regulation of Organ and Tissue Growth*, Academic Press, New York, 1972, p. 61.
7. Drachman, D. B. and Smith, A. L., *Trophic Functions of the Neuron, Ann. N.Y. Acad. Sci.*, 1973, 228, 1.
8. Guth, L., Watson, P. K. and Brown, W. C., *Exptl. Neurol.*, 1968, 20, 52.
9. —, and —, *Ibid.*, 1967, 17, 107.
10. Kuffler, S. W. and Vaughan-Williams, E. M., *J. Physiol.* (London), 1953, 121, 589.
11. Cavanaugh, G. M., *Formulae and Methods*, Marine Biological Laboratory, Woodshole, Mass., 1956, Vol. 4.
12. Bojji Reddy, N. and Swami, K. S., *Ind. J. Exptl. Biol.*, 1971, 9, 32.
13. Davis, B. J. and Orstein, L., *Disc Electrophoresis*, Distillation Products Industries, Rochester, U.S.A., 1961.
14. Hajek, I., Gutmann, E., Klitpera, M. and Syrory, I., *Physiol. Bohemoslov.*, 1966, 15, 148.
15. Sreelakshmi, P. and Swami, K. S., *Enzymologia*, 1972, 43, 373.
16. Morgulis, S. and Osherhoff, W., *J. Biol. Chem.*, 1938, 124, 767.

SHORT TERM GEO-STIMULATION AND P^{32} DISTRIBUTION IN ALASKA PEA SEEDLINGS

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PLANTS exhibit two growth responses—geotropism and gravimorphism, when their 'preferred' orientation with respect to gravity is disturbed. A number of changes in the cell and tissues probably underlie these responses.

Auxin moves to the lower side of the stimulated organ[1, 2] and increases the extensibility of the lower epidermal cells[3] with a concomittant enhancement of cellulase activity[4]. Induction of differences in the electrical potential[5] and redistribution of growth inhibitors[6] on the upper and lower side of the stimulated organs as well as the increase in the ethylene production, resulting in the leaf epinasty of stimulated shoots[7] have recently been reported. In the cells of the geo-stimulated organs, the amyloplasts[8,9] and Golgi Apparatus[10] move downward in the direction of gravity.

Increase in the weight of the lower leaves[11] and the lower half[4] of the horizontal stem strongly suggests the movement of food material such as reducing sugars[12] in the direction of gravity. The same effect of geo-stimulation may be expected for the inorganic ions, however, the data are insufficient in this regard. In one instance, the changes in the contents of certain ions have been studied in variously oriented young pine shoots[13].

For this reason, the distribution of leaf-fed P^{32} has been studied in geo-stimulated 3-leaf Alaska pea seedlings, over a short period of time.

The seedlings were raised under controlled conditions[14], in vermiculite, irrigated with Hoagland solution. Seedlings of uniform size were selected and divided into four groups: the plants of group A were retained in normal vertical orientation as control; the plants of group B were inverted; and those of groups C and D were laid horizontally and held in their respective orientations with the help of 'Twistems', leaving the roots undisturbed. Leaf-2 was below the axis in group C and above the axis in group D (Fig. 1). Ten microcurie of P^{32} as phosphoric acid in ten microliter drop of $0 \cdot 2$ M Na_2HPO_4 was applied to a leaf-let of leaf-2. The plants were harvested six hours after both the treatments according to the protocol given in Fig. 1. Radioactivity of the samples was counted and the data were processed as in an earlier paper[14]. Statistical comparisons of various parts of groups A and B, B and C, C and D, D and A, A and C, A and (C + D) and B and (C + D) were made by applying t-test.

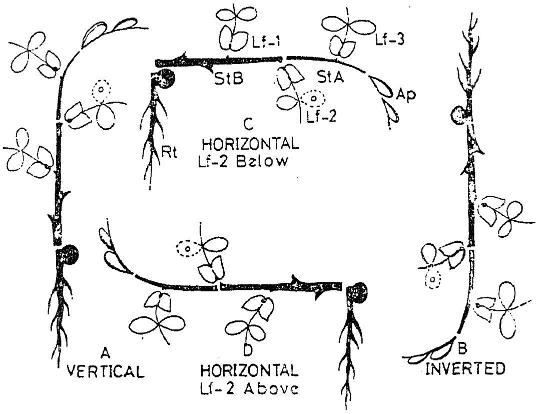

FIG. 1. Structure, orientations and harvest protocol of 3-leaf Alaska pea seedlings. Notations: Ap, Stem Apex; StA, Stem-above, StB, Stem-below, Rt, Root + Cotyledons and Lf, Leaf. P^{32} was applied to Lf-2. The circle in the dotted leaf-let represents the site of treatment. This leaf-let was severed and discarded six hours after treatment.

It was observed that the stem apex of the horizontal plant re-oriented itself, due to the curvature of the subtending fifth internode, within two hours of geo-stimulation.

It can be seen from Table I that greater the degree of disturbance in orientation the greater was the recovery of P^{32} from the seedlings. Since the recovery of P^{32} was the direct result of its absorption, it is inferred that geo-stimulation enhanced P^{32} absorption. The reason for this enhancement is not understood.

The distribution of P^{32} in various parts of the seedling is shown in Table II *a* and *b*. It may be noted that in contrast to the control and horizontal plants, the stem apex of the inverted seedling showed largest accumulation of P^{32}, both in terms of percentage and specific activity. This strongly suggests the movement of isotope in the direction of gravity. But in stem-above, in this treatment accumulated less P^{32} as compared to its counterpart in the control. In horizontal orientations, however, the same part accumulated more P^{32} in position D than in position C, because in the former the stem-above was slanting upward and in the latter downward.

The treated leaf-2 of the control plant, retained more but statistically insignificant amount of P^{32} as compared to its counterpart in the inverted plant (Table II *a*). On the contrary, in terms of specific activity (Table II *b*), this leaf of inverted plant retained more isotope than that of control.

The stem-below is basically a non-growing part. It conducts P^{32} either to stem apex or to roots. Thus the amount of isotope in this part depends upon the efficiency of the 'sinks' elsewhere. This part accumulated more P^{32} in orientation B than in A and more in orientation C than in D.

The roots of inverted plants—group B, accumulated more P^{32} as compared to those of vertical plants—group A, a situation contrary to the stem apex. This suggests that the response of the geo-stimulated root differs from that of shoot with regard to phosphate accumulation. This conclusion is further supported by the fact that P^{32} accumulation in the roots of the plants of groups A, C and D was not statistically different. The roots in these treatments were in natural orientation, whereas the shoots of C and D were geo-stimulated.

TABLE I

Recovery of p^{32} from 3-leaf Alaska pea seedlings in various orientations

Orientations			
Vertical (A) (n = 12)	Inverted (B) (n = 9)	Lf–2 below the axis (n = 13)	Horizontal (C & D) Lf–2 above the axis (n = 13)
Count Rate ± Standard Deviation (CPM)			
20918±5116 *ab* ac* a (c + d)**	37752±19522 *ab**	34322±21641 *ac* a (c + d)**	31055±11694 *a (c + d)**

The significant compared pairs are indicated by corresponding small letters.
* $p < 0{\cdot}05$.

TABLE II

Accumulation of P^{32} in different parts of variously oriented 3-leaf Alaska pea seedlings

Plant parts	Orientations A	B	C	D
	a. Percentages + Standard Deviation			
Ap	18·95±6·39 *ab**	24·24±4·02 *ab* bc**	18·21±4·65 *bc***	21·19±5·67
StA	34·45±7·28 *ab**	27·88±4·62 *ab* b (c + d)**	30·55±5·42 *b (c + d)* cd**	36·51±9·42 *b (c + d)* cd**
Lf–2	5·15±1·03	4·54±0·76 *bc**	7·28±4·73 *bc**	5·80±3·33
StB	22·76±5·76	21·34±5·08	26·41±7·47 *cd***	19·79±5·66 *cd***
Rt	18·69±6·78	22·00±3·96 *b (c + d)**	17·55±4·75 *b (c + d)**	15·71±6·61 *b (c + d)**
	b. Specific activities (CPM/mg dry weight) ± Standard Deviation			
Ap	541±243 *ab** da**	1193±244 *ab** bc* b (c + d)**	748±616 *bc* b (c + d)**	922±518 *da* b (c + d)**
StA	303±112	425±47	447±318	442±179
Lf–2	175±58 ← *a (c + d)***	276±29 *bc***	473±247 *bc** cd** a (c + d)***	279±142 *cd** a (c + d)***
StB	140±40 *ab* a (c + d)*	217±52 *ab**	246±100 *cd* a (c + d)***	183±89 *cd* a (c + d)***
Rt	54±16 *ab***	115±41 *ab***	85±57	77±57

The significant compared pairs are indicated by corresponding small letters.
* $p < 0{\cdot}05$; ** $p < 0{\cdot}01$.

In horizontal plants-groups C and D, when leaf-2 was above the axis, the other two leaves were below the axis, or *vice versa*. Leaf-1 was a part of the stem-below and leaf-3 of the stem-above. To illustrate the effect of geo-stimulation further, the dry weights and P^{32} accumulations of leaves-1 and -3 have been summed up in Table III. The data clearly show that the dry weight of these and leaf-2

TABLE III

Dry weights and P^{32} accumulation of leaves in above or below the axis positions in horizontally oriented (C and D) seedlings

Leaves	Above the axis	Below the axis
I. Dry weights (Milligrams)		
(1+3)	16·7	22·5
2	6·0	6·2
II. P^{32} Accumulation		
a. Percentage ± Standard deviation		
(1+3)	0·38±0·40 *cd**	1·06±1·04 *cd**
2	5·80±3·33	7·28±4·73
b. Counts/minute ± standard deviation		
(1+3)	122±139 *cd**	393±356 *cd**
2	1700±833 *cd**	3448±584 *cd**
c. Specific activity ± standard devistion		
(1+3)	7±2 *cd**	17±16 *cd**
2	279±142 *cd**	556±247 *cd**

The significant compared pairs are indicated by the corresponding small letters.

* $p < 0·05$.

increased in below-axis position and decreased in above-axis position, confirming earlier reports[11,13]. The accumulation of P^{32} followed the same relationship, indicating the movement of isotope and probably other food materials in the direction of gravity. However, the movement of P^{32} also occurred horizontally, resulting in the isotope distribution in the whole plant, similar to the movement of other inorganic ions in young pine shoots[13].

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1. Gillespie, B. and Thimann, K. V., *Plant Physiol.*, 1963, 38, 214.
2. Ishii, S., *Proc. Crop. Sci. Soc. Jap.*, 1972, 41, 22.
3. Iwami, S. and Masuda, Y., *Plant and Cell Physiol.*, 1974, 15, 121.
4. Bara, M. and Gordon, S. A., *Physiol. Plant.*, 1972, 27, 277.
5. Etherton, B. and Dedolph, R. R., *Plant Physiol*,, 1972, 49, 1019.
6. Borkowska, B., *Acta Agrobot.*, 1973, 26, 161.
7. Leather, G. R., Forrence, L. E. and Abelle, F. B., *Plant Physiol.*, 1972, 49, 183.
8. Griffiths, H. J. and Audus, L. J., *New Phytol.*, 1964, 63, 319.
9. Shen-Miller, J. and Miller, C., *Plant Physiol.*, 1972, 49, 634.
10. Juniper, B. E. and French, A., *Planta*, 1970, 95, 314.
11. Ghent, A. W., *New Phytol.*, 1973, p. 1141.
12. Bridges, I. G. and Wilkins, M. B., *Planta*, 1974, 117, 243.
13. Murtov, Y. M. and Mitrifanov, D. P., *Fiziol Rast*, 1973, 20, 1062.
14. Husain, S. M. and Linck, A. J., *Physiol. Plant.*, 1966, 19, 992.

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LETTERS TO THE EDITOR

VANILLYDENE-*o*-TOLUIDINE COMPLEXES OF BIVALENT COBALT, MANGANESE, ZINC AND OXOVANADIUM(IV)

Introduction

In continuation of our earlier work on the bivalent transition metal complexes with the Schiff bases derived from vanillin[1], we now report the complexes of cobalt(II), manganese(II), zinc(II) and oxovanadium(IV) with vanillydene-*o*-toluidine.

TABLE I

S. No.	Elemental analysis* %				Infrared frequencies cm^{-1}					Electronic spectra† nm
	Empirical formulae	N	Cl	Metal	νOH	νC=N	νC=C	νC—O	νV=O	Absorption bands and intensities $\epsilon \times 10^{-4}$
1.	$C_{15}H_{15}O_2N$ (Ligand)	..	..	..	3500	1630	1605 1590 1[illegible]40 1510	1290 1265	..	230 (0·40); 250(0·61); 280(1·09); 310(1·16); 330 (1·11): 350 (0·93)
2.	$(C_{15}H_{15}O_2N)\,CoCl_2$	3·67 (3·76)	18·80 (18·82)	16·10 (16·40)	3400 3160	1635	1590 1560 1505 1500	1310	..	250(1·34); 280(1·66); 325(2·12); 610(0·025); 675(0 038)
3.	$(C_{15}H_{15}O_2N)\,MnCl_2$	3·45 (3·80)	18·76 (19·02)	14·70 (14·97)	3310 3100	1635	1590 1570 1550 1505 1500	1325	..	220 (0·91); 250 (1·19); 280 (1·51); 320 (1·78)
4.	$(C_{15}H_{15}O_2N)\,ZnCl_2$	3·42 (3·70)	18·23 (18·49)	17·12 (17·33)	3400 3160	1635	1590 1550 1510 1500	1320	..	250 (1·49); 295 (1·77); 320 (2·34)
5.	$(C_{15}H_{15}O_2N)\,VOCl_2$	3·35 (3·69)	18·52 (18·74)	13·11 (13·46)	3400	1630	1580 1510	1285	990	250 (0·94); 270 (1·35); 310 (1·08); 590 (0·053); 920 (0·048)

* The calculated values are shown in the parenthesis. † The intensities are given in the parenthesis.

Experimental

The chemicals used were of high purity grade. Metal chlorides used for preparing the complexes were analar BDH reagents. Vanadyl chloride was a BDH reagent.

Preparation of the Ligand.—Vanillin and *o*-toluidine in stoichlometric quantities (1 : 1) were refluxed in ethanol for about two hours. The Schiff base which separated on cooling was recrystallised from ethanol and dried in vacuum over fused calcium chloride.

Preparation of the Complex.—Vanillin, *o*-toluidine and the corresponding metal chlorides in appropriate molar ratio (1·2 : 1·2 : 1) were refluxed for two to three hours with ethanol. The precipitated complexes were filtered, washed with dry ethanol and dried in vacuum over fused calcium chloride. The complexes were further purified by subjecting them to soxhlet extraction using dry ethanol.

Analysis.—The metals, nitrogen and chloride were estimated by standard procedures[2]. The results of analysis are shown in Table I.

Physical Measurements.—The electronic spectral measurements were made on Hilger Uvispeck Spectrophotometer model H700, using one cm matched quartz cells. The ELICO bridge (cell constant 0·829) was used for conductivity measurements. In both the cases, DMF was employed as the solvent. The infrared spectra of the ligand and the complexes were recorded on a Perkin-Elmer Spectrophotometer-221, using KBr pellet.

Results and Discussion

The complexes have 1 : 1 stoichiometry and are soluble in DMF and DMSO.

Conductance Measurements.—The molar conductance values in DMF at the concentration 10^{-1} M show no appreciable dissociation suggesting non-electrolyte behaviour[3].

Electronic Spectra.—The electronic spectra of the ligand and the complexes are very similar in the 210–390 nm region apart from the enhanced intensities in the complexes. The intensity values given in Table I reflect this.

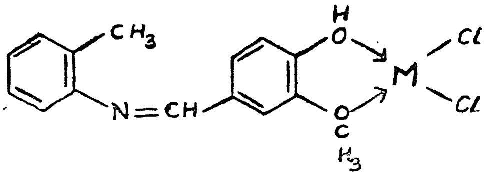
Cobalt (II) complexes show an absorption spectrum (λ, 610 nm, ϵ, $2{\cdot}549 \times 10^2$: λ, 670 nm; ϵ, $3{\cdot}84 \times 10^2$) indicative of tetrahedral nature of the complex.

The visible spectrum of oxovanadium (IV) complex can be interpreted in terms of C_{4v} symmetry by attributing the prominent band at 920 nm to the $d_{xy}-d_{yx}$, d_{xz} transition and the other at 590 nm in the form of a shoulder to the $d_{xy}-d_{x^2-y^2}$ transition. The charge transfer band in the region 320–350 nm appearing as a tail of high intensity is assigned to $d_{xy}-d_{z^2}$ transition. These assignments are in agreement with the earlier reported values[4].

Infrared Spectra.—The main infrared frequencies and their assignments[1,6,7] in the ligand and the complexes are given in Table I.

The lowering of the intense band at 3500 cm^{-1} in the free ligand to 3400–3300 cm^{-1} in the complexes is indicative of coordination through the OH group of the ligand. The apparent constancy of the band at 1635–1630 cm^{-1} in the ligand as well as the complexes suggest nonparticipation of nitrogen of C=N in the coordination to the metal. Also the phenolic C—O frequency in the ligand is moved to a higher frequency in the complexes showing involvement of this group in the coordination to metal.

Thus the situation in the complexes can be visualised as follows:



The 990 cm^{-1} band in the oxovanadium (IV) complex suggests the considerable π character of V—O bond and the monomeric nature of the complex[5].

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1. Pujar, M. A., Hiremath, A. C. and Prabhakar. B. K., *J. Indian Chem. Soc.*, 1974, 51 (4), 494.
2. Vogel, A. I., *A Text Book of Inorganic Analysis*, Longmans Green and Co. Ltd., London, 1962.
3. Quagliano, J. V., Fujita, J., Franz, G., Philips, J. D. J., Walsmly, J. A. and Tyree, S. Y., *J. Am. Chem. Soc.*, 1961, 83, 3770.
4. Biradar, N. S., Miss Devale, S. and Kulkarni, V. H., *Curr. Sci.*, 1973, 42 (4), 125 and references therein.
5. Sathyanarayana, D. N. and Patel, C. C., *J. Inorg. Nucl. Chem.*, 1966, 28 2277 ; *J. Sci. Ind. Res.*, India, 1968, 27, 348.
6. Freedman, H. H., *J. Am. Chem. Soc.*, 1961, 83, 2900.
7. Kovacic, J. E., *Spectrochim. Acta*, 1967, 23 A, 183.

OXIDATION OF CROTYL ALCOHOL WITH CHLORAMINE-T

During some of our investigations[1-3] on the oxidation of alcohols by chloramine-T, it was found that crotyl alcohol undergoes oxidation with a two electron change, with chloramine-T (CAT). A survey of literature shows little information on its oxidative behaviour or on analytical techniques for its estimation. The present communication reports an elegant method for the estimation of crotyl alcohol in aqueous solutions, using CAT. Although the oxidation was rapid, it was not instantaneous and hence a back titration procedure was employed.

Materials and Methods

Crotyl alcohol (Fluka, b.p. 125° C, $n_D = 1{\cdot}424$) was used without further purification. A weighed quantity of the alcohol was dissolved in the appropriate solvent to give an approximately 0·2 M solution. CAT was purified by the method of Morris *et al.*[4]. An approximately decinormal solution was prepared and standardized by the iodometric method. All other reagents were of accepted grades of purity. Standard buffer systems were employed. Triply distilled water was used for preparing the solutions.

Preliminary Studies

Known amounts of crotyl alcohol solution in the proper solvent were added to a known excess of CAT in an iodine flask at room temperature (25 ± 2° C). The reaction mixture was kept aside for various intervals of time and then the excess of CAT determined by back titration. A period of 10 minutes was adequate for the quantitative reaction in certain solvents. A typical set of results for the oxidation of the alcohol by CAT is given in Table I.

TABLE I

Oxidation of crotyl alcohol with chloramine-T in 10 min. at various pH values

Medium	Ratio of $\frac{\text{CAT used}}{\text{alcohol taken}}$
0·1 N HCl	0·984
0·2 N HCl	1·012
0·3 N HCl	1·018
0·4 N HCl	1·018
0·5 N HCl	1·021
1·0 N HCl	1·023
1·5 N HCl	1·033
*pH 1·0	1·045
*pH 2·0	1·016
pH 3·0	1·006
pH 4·0	1·021
pH 4·2	1·022
pH 5·0	1·023
pH 6·2	0·923
pH 7·0	0·783
0·1 N NaOH	0·018
0·1 N NaOH + OsO_4	0·101
0·5 M $HClO_4$	0·349
0·5 M H_2SO_4	0·307

* HCl–KCl buffer.
pH 3–7: Citric acid—Phosphate buffers.
crotyl alcohol: 0·5 m. mole, CAT taken, 4·5 m. mole.

It is seen that oxidation is rapid and stoichiometric in HCl medium (0·2 — 0·4 M) and in buffer media of pH 1–5, but is slow in presence of sulphuric and perchloric acids and at pH greater than 5. However, if the reaction is carried out in 0·5–1·0 M HCl stoichiometric oxidation occurs within 2–3 minutes. It was also noted that substantial amounts of the alcohol can be estimated, within 10 minutes, by having a 10–20% excess of the oxident in solution.

Recommended Procedure

Add aliquots of an aqueous solution of crotyl alcohol to 25 ml of 0·1 N CAT containing enough 2 M HCl to give an overall acid concentration 0·5–1·0 M. Shake the mixture. After 5 minutes add 20 ml of 10% potassium iodide and titrate with decinormal sodium thiosulphate. A blank is run with CAT solution alone. The amount of crotyl alcohol in the experimental solution is calculated on the basis of equation (1).

Results and Discussion

The stoichiometry of the oxidation can be represented as $CH_3CH = CHCH_2OH + RNClNa \rightarrow CH_3CH = CHCHO + RNH_2 + NaCl$ ... (1)

where $R = p - CH_3C_6H_4SO_2$.

The presence of crotyl aldehyde in the reaction products was shown by spot tests[5]. Further, the aldehyde was isolated as its 2–4 dinitrophenyl hydrazone[6] (MP. 189° C) and paper chromatography was used to identify the sulphonamide[7] ($R_f = 0{\cdot}905$). The stoichiometry is unaffected by a reversal of the order of addition of the oxidant and crotyl alcohol. Also neutral salts have no influence on the rate of oxidation and stoichiometry, but the presence of perchloric or sulphuric acid considerably retards the reaction. Kinetic investigations are in progress and the latter could probably be attributed to a specific inhibitory effect of ClO_4^- and SO_4^{2-} ions. Similar observations have been made in the oxidation of allyl[1] and cinnamyl alcohols[2] with CAT. Some typical results of analysis are given in Table II. The values obtained are accurate within 0·5%.

TABLE II

Estimation of crotyl alcohol with chloramine-T

Alcohol taken (mg)	Alcohol found (mg)
3·99	4·01
7·99	8·00
10·48	10·46
13·98	13·99
17·47	17·45
20·96	20·99
24·46	24·67
27·95	27·97
31·05	31·06
34·94	34·95
38·93	38·74

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1. Mahadevappa, D. S. and Naidu, H. M. K., *Talanta,* 1973, 20, 349.
2. — and —, *Indian J. Chem.,* 1975, 13, 976.
3. — and —, *Australian J. Chem.,* 1974, 27, 1203; 1975, 28, 899.
4. Morris, J. C., Salazar, J. A. and Wineman, M. A., *J. Amer. Chem. Soc.,* 1948, 70, 2036.
5. Feigl, F., *Spot Tests in Organic Analysis,* Elsevier, Amsterdam, 1956, p. 224.
6. Cheronis, N. D., Entrikin, J. B. and Hodnett, E. M., *Semi Micro Qualitative Organic Analysis,* (Interscience, New York), 1965, p. 728.
7. Mahadevappa, D. S. and Gowda, N. M. M. *Talanta,* 1975, 22, 771.

SPECTROPHOTOMETRIC STUDIES ON Zn^{+2} WITH 7-α-ANILINO BENZYL-8-QUINOLINOL (ABQ)

7-α-Anilino benzyl 8-Quinolinol (ABQ) was first prepared by Pirrone[1] and Phillips[2] and its chelating tendency was first qualitatively studied by Phillips[3]. A survey of literature reveals that its stability and spectrophotometric study has not been studied. This study was undertaken by us. The stability, composition and its use as spectrophotometric reagent for bivalent metal Zn (II) chelate of ABQ has been determined spectrophotometrically in 60% *v/v* dioxane : water system using Job's Continuous Variations, molar ratio and slope ratio methods.

Experimental

The $ZnSO_4$ solution was standardised by EDTA, titration and was further diluted to provide 4×10^{-3} M Zn^{+2} solution. Ligand ABQ (4×10^{-3} M) solution was prepared in pure dioxane. Buffer solutions had a pH range of 0·65 to 9·6. The absorbance readings were taken on Bosch-Lomb spectrophotometer at 30 ± 1° in 60% *v/v* dioxane : water. The spectral study showed λ_{max} at 390 nm at pH 7·6.

The nature of the complex was studied by employing three different methods, namely, Voxburgh and Cooper[4], Job's method of Continuous Variations[5] using equimolar solutions Slope ratio[6] and Molar ratio[7].

Job's method of continuous variations.—For this measurement 4×10^{-4} M solutions were employed at 7·6 pH, the absorption being measured at 390 nm. The results showed maximum absorption at 0·33 mole fraction which indicates the formula of the chelate as 1 : 2 [Zn (ABQ)$_2$]. Slope ratio and Molar ratio also gave similiar results. The stability constant of the complex as calculated by Dey and Banerjee's Method[8], was $6 \cdot 2 \times 10^{13}$. The free energy of formation was 26·8 K. cals/mole at 30° ± 1° C. The mole ratio method gave stability constant of $4 \cdot 0 \times 10^{13}$. The method is useful for spectrophotometric determination of Zn^{+2} by ABQ at 4–8 pH.

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1. Pirrone, F., *Gazz. chim. ital.,* 1940, 70, 520.
2. Phillips, J. P., Keown, R. and Fernando, Q., *J. Amer. Chem. Soc.,* 1953, 74, 4306.
3. —, — and —, *Ibid.,* 1955. 77, 5504.
4. Vosburgh, W. C. and Cooper, G. R., *Ibid.,* 1941, 63, 437.
5. Job, P., *Ann Chim.,* 1928, 9, 113.
6. Harvey, A. E. and Manning, D. L., *J. Amer. Chem. Soc.,* 1950, 72, 4448.
7. Yoe, J. H. and Jones, A. L., *Ind. Eng. Chem. Anal. Ed.,* 1944, 16, 111.
8. Banerjee, S. K. and Dey, A. K., *Z. Anal. Chem.,* 1961, 179 (1), 30.

SPECTROPHOTOMETRIC DETERMINATION OF RUTHENIUM (III) WITH THIORIDAZINE HYDROCHLORIDE

Introduction

REAGENTS proposed for the spectrophotometric determination of ruthenium have been reviewed[1-5]. Thioridazine hydrochloride (TH) recommended by the authors for the determination of palladium (II)[6] has now been used for the spectrophotometric determination of ruthenium (III).

Experimental

Standard solution of ruthenium (III) was prepared from ruthenium (III) chloride in 1·0 N hydrochloric acid and standardised. A 0·2% aqueous solution of TH was used. All other solutions were prepared from reagent grade chemicals. Beckman Model DB Spectrophotometer was used for absorbance measurements.

Procedure.—Transfer an aliquot of the sample solution which contains from 20 to 200 μg of Ru(III) to a 25 ml volumetric flask. Add sufficient sulphuric acid so that the final solution will be 1·5 N in sulphuric acid. Add 5 ml of 0·2% TH. Dilute to volume with double distilled water. Measure the absorbance *vs* a reagent blank at 640 nm after 10 minutes. Compare the absorbance with a standard curve to calculate the amount of ruthenium (III).

Results and Discussion

TH forms a blue coloured species with ruthenium(III) instantaneously at room temperature (22°–25° C) in sulphuric or hydrochloric acid. The sensitivity in the four acid media (1·5 N) is in the order—HCl > H_2SO_4 > H_3PO_4 > HAC. The maximum colour development takes place in 10 minutes after mixing the reagents. The stability of the blue colour in 1·5 N H_2SO_4, HCl, H_3PO_4 and HAC is 30, 20, 8 and 5 minutes respectively. Hence sulphuric acid medium has been selected. Nitric acid medium cannot be used as it oxidises TH.

The maximum colour intensity is observed in 0·1–2·5 N H_2SO_4. Above 3·0 N precipitation of the reagent takes place. A fifteen-fold molar excess of the reagent was necessary for the full development of the colour intensity. The blue coloured species assumed to be a radical exhibits absorption maximum at 640 nm at which the reagent does not absorb. The absorbance values remained constant for 30 minutes and were insensitive to temperature in the range 5–50° C. The order of addition of reagents was not critical. Beer's law is obeyed from 0·8 to 8·0 ppm of ruthenium. The optimum concentration range evaluated by Ringbom's method[7,8] is 1·5 to 7·0 ppm. The Sandell sensitivity of the colour system was found to be 0·0222 μg Ru(III) cm^{-2}. The molar absorptivity was $4 \cdot 58 \times 10^3$ litre $mole^{-1}$ cm^{-1}.

With the help of this reagent, as little as 4 ppm of Ru(III) can be determined in presence of 1000 ppm of the ions of Zn(II), Mg(II), Ca(II), Ba(II), Sr(II), fluoride, chloride, bromide, nitrate, sulphate, phosphate, oxalate, acetate; 100 ppm of Ni(II), Co(II), Cu(II), UO_2(II) and EDTA. However the ions of other platinum metals, Au(III), Ag (I), V(V), Ce(IV), thiosulphate and iodide were found to interfere at all concentrations.

Analysis of synthetic samples.—In one phase of the processing system devised for experimental Breeder reactor-II (EBR-II), molten zinc and zinc-magnesium alloy are used as solvents in the separation of uranium from ruthenium and other fission products. The results of analysis of ruthenium in the synthetic mixtures corresponding to zinc-magnesium alloy composition are given in Table I.

TABLE I

Determination of ruthenium in zinc-magnesium alloy

Zinc added, mg	Magnesium added, mg	Ruthenium added μg	Ruthenium found av. μg	No. of determinations
30	..	4·00	3·98	5
..	30	4·00	3·99	5
15	15	4·00	3·97	5
15	15	2·00	1·99	5

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1. Beamish, F. E. and Van Loon, J. C., *Recent Advances in the Analytical Chemistry of the Noble Metals,* Pergamon Press, Oxford, 1972, p. 375.
2. —, *Analytical Chemistry of the Noble Metals,* Pergamon Press, Oxford, 1966, p. 366.
3. Boltz, D. F. and Mellon, M. G., *Anal Chem.,* 1974, 46, 234 R; 1972, 44, 300 R; 1970, 42, 152 R; 1968, 40, 255 R; 1966, 38, 317 R.
4. Beamish, F. E. and McBryde, W. A. E., *Anal. Chim. Acta,* 1958, 18, 551; 1953, 9, 349.
5. —, *Talanta,* 1965, 12, 789.
6. Sanke Gowda, H., Ramappa, P. G. and Narayana Achar, B., *Z. anal. Chem.,* 1975, 275, 127.
7. Ringbom, A., *Ibid.,* 1938, 115, 332.
8. Ayres, G. H., *Anal. Chem.,* 1949, 21, 652.

THE EFFECT OF ULTRASOUND ON THE CATALYTIC ACTIVITY OF COPPER

THE reported work-hardening of fcc metals[1] and a 30–100 fold increase in the rate of recrystallisation, in oxygen-free, high-conductivity copper[2], both induced ultrasonically, and the variation of the velocity of ultrasound and attenuation in copper under varying static stress[3], when viewed in the light of Hutchison's review[4] on ultrasonic absorption in solids, clearly indicate the existence of a definite interaction between ultrasound and dislocations in a solid. If the ultrasound can also bring about an alteration in the catalytic activity of a metal, that will, indeed, provide a new evidence in favour of Cratty and Granato's hypothesis[5] that the surface terminations of dislocations are one type of active centres. A survey of the literature does not reveal any attempt made in this direction, hence, the present investigation.

Square pieces (3·0 cm × 3·0 cm) of copper foils (99·9%, B.D.H.) were used; the gm. equiv. of hydrogen peroxide (0·25 N) decomposed per unit surface area of the foil during the initial 10 min. of the reaction at 69·5° C at a stirrer speed of 200 rpm was taken as a measure of the activity, *a*, of the foil.

In one set of exploratory experiments the copper foils were first annealed in air for one hour at $T_A = 150°–480°$ C and then subjected to the following sequence of treatments:

I (*a*) [QD], I (*b*) [QID], I (*c*) [QDID] where

Q, quenching the annealed foil in water at 25° C; D, dipping the foil in 2 N H_2SO_4 for 3–4 min. until it regained its shine, *i.e.*, the surface oxide dissolved out; I, insonating the foil at frequency 80 kHz (60 W/cm^2) for 30 min. at 25° C. The apparatus for insonation consisted of an ultrasonic cleaner (Type 1201, Systronics) fitted with an electrical-to-mechanical power transducer at the bottom of the tank; the foil was placed in contact with the transducer with water (at 25° C) standing to a height of 9 cm above it; as there was warming up of the water during insonation, it was carried out with interruptions.

In another set of experiments the unannealed copper foils were immersed in 0·25 N hydrogen peroxide for 20 min. at 69·5° C to oxidise the surface and then subjected to either [ID], II (*a*) or [DID], II (*b*) treatment.

Figure 1 shows the variations of (a/a) with T_A, a_0 being the catalytic activity of the untreated foil ($1·14 \times 10^{-4}$ gm. equiv. of hydrogen peroxide decomposed cm^{-2} in 10 min. at 69·5° C).

The results of

I (*a*) simply confirm the earlier findings of the authors[6].

I (*b*) and I (*c*), though different from those of I (*a*), are new and significant; they clearly reveal that while in the presence of surface oxide, ultrasonic annealing of activity does occur, in the absence of the surface oxide, however, the activity, apparently, remains unchanged.

II (*a*) and II (*b*) showed that even without previous annealing the oxide-coated foils lost about 30% of their activity (a_0) while the 'oxide-free' ones lost no activity.

Incidentally, the observed absence of activity decay at $T_A < 200°$ C can, now, be attributed to negligible or partial oxidation of the metal at low T_A. [*see* curve I (*b*) in Fig. 1].

This observation assumes special significance when it is recalled that surface oxide has also been found to affect such of the bulk properties as internal friction[7] and steady state creep rate[8] of copper and other metals. Thus, it is clear that the effect of ultrasound on the catalytic activity of copper is independent of T_A but, dependent on the coverage of the metal surface by its oxide, no matter how the surface oxidation is brought about; a detailed correlation between this effect and the extent of surface oxidation is under investigation.

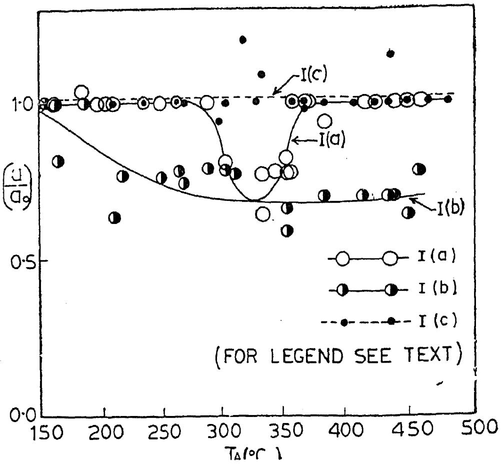

FIG. 1.

Anyway, the present investigation succeeds in providing not only a new and strong evidence in favour of Cratty and Granato's hypothesis[5], but also, a new insight into the possible relationship between the problem of active centres (on the surface of a metal catalyst) and such of the physical properties as internal friction, creep, recrystallisation, hardness, etc. Further studies are in progress.

The authors are thankful to the Director of the National Institute of Oceanography, Panaji, for making available the ultrasonic and other facilities.

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* This communication is based on a part of the research work carried out by K. S., for the Ph.D. degree of the University of Bombay.

1. Karlik, G. and Weiss, B., *Z. Metallk.*, 1967, 58, 471.
2. Hayes, G. A. and Shyne, J. C., *Metal Science J.*, 1971, 5, 19.
3. Alers, G. A. and Salama, K., *Dislocation Dynamics, Materials Science and Engineering Series*, McGraw-Hill Book, Co., New York, 1968, p. 211.
4. Hutchison, T. S., *Science*, 1960, 132, 643.
5. Cratty, L. E. and Granato, A. V., *J. Chem. Phys.*, 1957, 26, 96.
6. Sawkar, K. and Bhakta, M. A., *Curr. Sci.*, 1975, 44, 261.
7. Dhar, P. R. and Bhattacharjee, K. K. and Ghosh, S. S., *J. Sci. Ind. Res. (India)*, 1961, 20 D, 237.
8. Schröder, K., Giannuzzi, A. J. and Gorsha, G., *Acta Met.*, 1968, 16, 469.

OCCURRENCE OF THE TRACE FOSSIL *HIRMERIA* FUCINI 1936, IN THE INTERTRAPPEAN BED OF NINAMA, SURENDRANAGAR DISTRICT, GUJARAT

WE wish to report the first occurrence of a trace fossil from the Intertrappean Bed at Ninama (22° 18′ : 71° 20′) which adds siginificantly to our knowledge about the depositional conditions of this bed.

By its narrow sharp crested ridges running subparallel and criss-crossed by similarly narrow canals, to produce an intricate pattern, the present material can be easily identified with *Hirmeria khadluensis* Chiplonkar and Ghare[2] reported from the cherty limestone of the Lameta Beds around Khadlu, 2 km south of Mongra (22° 00′ 38″ : 74° 02′ 30″) ; the only observable difference is that the ridges in the specimens from Khadlu are slightly less sharp crested than in the present material.

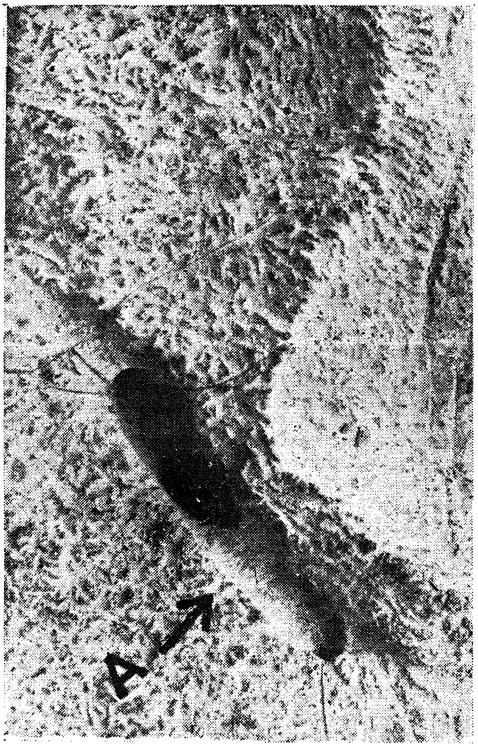

FIG. 1. *Hirmeria khadluensis* Chiplonkar and Ghare, × 1·5—"A" Associated burrow fossil.

Hirmeria Fucini is interpreted[3] as due to inorganic process and its occurrence along with a burrrow fossil (indicated by "A" in Fig. 1) belonging to the *Domichnia* type of Seilacher's[4] ethological group, indicates a period of retardation in deposition and noneuxinic origin for the sediment.

The burrow ("A" in Fig. 1), associated with the present trace fossil, is inclined to the bedding plane at a very low angle, suggestive of the animal moving in just sub-surface position, and, collecting food from the sediment surface where it was accumulating under conditions of retarded sedimentation.

While *Hirmeria* Fucini, 1936, comes from (?) Lower Permian, of Itali (Verra Cano), the occurrences of *Hirmeria* in India are from cherty limestones of fresh water lacustrine deposits (Lameta and Intertrappean Beds) of much higher horizons, *viz.*, late Creteaceous to Palaeocene as indicated by the conventional fossils[1,2].

Maharashtra Association G. W. CHIPLONKAR.
for the Cultivation of Science, R. M. BADVE.
Poona-4, *May* 24, 1975.

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1. Borkar, V. D., *Proc. Ind. Acad. Sci.*, 1973, **78 B** (4).
2. Chiplonkar, G. W. and Ghare, M. A., *Bull. Ind. Geol. Assoc.*, 1975, **8** (1).
3. Häntzschel, W., *Treat. Invert. Pal.*, Ed. Moore, R. C., 1962, W (Univ. Kansas Press).
4. Seilacher, A., In : *Approaches to Paleoecology*, Eds. Imbrie, J. and Newell, N. D. (John Wiley and Sons, Inc., New York), 1964.

A NOTE ON THE PSEUDO-OOLITES FROM THE GREAT LIMESTONE OF LAIN, UDHAMPUR DISTRICT, JAMMU PROVINCE, J AND K STATE (INDIA)

THE present note deals with the finding of pseudo-oolites from the Great Limestone of Lain (Lat. 32°. 59′ 30″ N and 75° 02′ 00″ E, 43p/1). The Great Limestone of Lain is the oldest formation in the area and its base is not exposed in the area covered. It is overlain by the Eocenes and other late Tertiary formations.

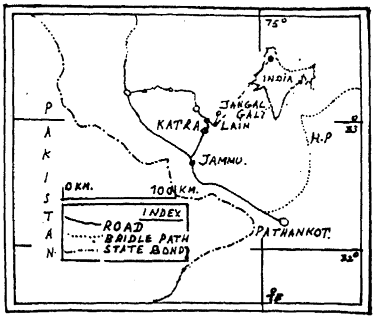

FIG. 1. Location Map.

The Great Limestone portrays variation both in physical and chemical traits within wide limits. At places, the limestone is partly or completely dolomitised. It is massive, hard, dense, usually non-crystalline extensively jointed, fissured and fractured in an irregular pattern. While making a sedimentological studies of the rocks of the area, the author came across some samples of the limestone which exhibited pseudo-oolites for the first time.

The pseudo-oolites are light grey to brown and dark brown in colour. The intensity of colour decreases from centre outward, towards the periphery. Areas in which the grains are exceedingly fine or impure appear dark, while those in which the grains are coarser and purer have a distinctly lighter shades. The fine grained pseudo-oolites are mostly oval in outline and are dark and embedded in a clearer and coarser calcite. The ovules may represent fecal pellets of organisms that lived in the muds when they were being deposited. The colouring pigment is generally of organic origin. The average size of the pseudo-oolites varies from 0·086 to 1·5 mm and they have no genetic relation with the number of rims or the nuclei, which ordinarily control the size of the normal oolites.

In a few sections, some of the pseudo-colites appear to be deformed. The deformation is due, either to corrosion *in situ,* contraction of certain individuals in a matrix which would permit a relatively easy movement of the materials. or due to strong agitation in the environments. Since the pseudo-oolites have no concern with agitated waters, as they are formed mostly in an eperic environment of quiet sedimentation, it appears plausible that the deformity was due to algal activity, at the time of sedimentation. The algae was blue green belonging to collenia group. The deformation resulted in perforation, punctuation and break up of the original set up of the pseudo-oolites.

The pseudo-oolites appear to have been formed in marine, shallow, quiet water environments. The algae played a vital role for the entrapment of precipitated carbonate.

The Great Limestone of Jammu has a close resemblance to that of the Krols, from which the algal oolites have been reported. The presence of fossil "Oncolite" (C. Gundu Rao, 1970) from the Krols has established their upper Pre-Cambrian age. It is thus obvious that the pseudo-oolites have thrown much light on the diagenetic history of the Great Limestone. The Great Limestone of Jammu from which the pseudo-oolites have been found now are of algal origin and are of an upper Pre-Cambrian age.

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---

1. Bassi, U. K. and Upendra, S. V., *Him. Geol.,* 1971, 1, 244.
2. Carrozzi, A. V., *Microscopic Sedimentary Petrography,* John Willey and Sons, Inc., New York, 1960, p. 238.
3. Freeman, T., *Journ. Sed. Petrol,* 1962, 1 (88), 475.
4. Medlicott, H. B., *Mem. G.S.I.,* 1864, **8**, Part 2. p. 1.
5. Middlemiss, C. S., *Min. Sury. Report,* J and K Govt., 1929.
6. Pettijohn, F. J., *Sedimentary Rocks* (2nd Edition), Oxford Book. Co., New Delhi, 1929, p. 94.
7. Rao, C. Gundu, *Pub. Cent. Adv. Geol.,* No. 7, October 1970, p. 127.
8. Sharma, M. R., *University Review* (J and K University), 1973, 3 (6), 15.
9. Simpson, R. R., *Mem. G.S.I.,* 1904, **32** (4), 194.
10. Williams, H., Turner, Francis, J. and Gilbert, Charles, M., *Petrography—an Introduction to the Study of Rocks in Thin Sections Vakils,* Feffer and Simons (P), Ltd, Bombay, p. 348.

DISCOVERY OF DEVONIAN SCOLECODONTS IN THE LESSER HIMALAYAN ZONE OF GARHWAL, U.P.

THE find of bryozoans, brachiopods and other fossils from the shaly horizon of the Boulder Slate Member forming part of the "Lower Bijni Tectonic Unit" exposed on the southern limb of the Garhwal Syncline has aroused much interest among those working on Himalayan Geology[1-3]. On the basis of fossils collected from different localities, Middle Carboniferous to Permian ages were assigned to the fossiliferous beds[2-4]. According to Ravi Shanker and Ganesan[5] the beds yielding Upper Palaeozoic fossils are conformably overlain by the gritty quartzite and sandy limestones.

One of us (B. S. T.) visited the Dogadda (29° 48′ 24″ : 78° 36′ 48″) area of Garhwal and collected rock specimens from different stratigraphic horizons. One of these samples, collected from a locality about 1·5 km north of Dogadda from the sandy limestone horizon on maceration in N/10 acetic acid has yielded fairly well preserved scolecodont assemblage (Fig. 1) of probable Devonian age. The fauna includes *Nereidavus* aff. *ontarioensis* Stauffer, *Nereidavus* aff. *planus* Stauffer, *Kettnerites* cf. *kosoviensis* Zebera, *Eunicites* sp. In addition to these, specimens of worm tubes referable to *Protoscolex* sp. were also obtained. All the scolecodont forms referred to, above have close affinity with Devonian scolecodonts.

The find of Devonian scolecodont fauna from the beds conformably lying above the Boulder Slate

Member and gritty quartzites of Upper Palaeozoic age is significant as it is suggestive of the sequence being inverted. The work on the fauna is in progress and the details will be published shortly.

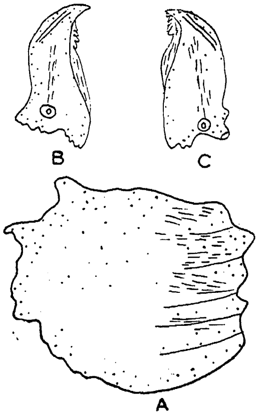

FIG. 1. A, *Kettnerites* cf. *kosoviensis* Žebra, × 80, dorsal view; B and C, *Nereidavus* aff. *ontarioensis* Stauffer, × 50, dorsal and ventral views.

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Panjab University,
Chandigarh, *December* 12, 1975.

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---

1. Ganesan, T. M., *Indian Minerals,* 1971, 25 (3), 257.
2. —, *Himalayan Geology,* 1972, 2, 431.
3. Ravi Shanker, Dhoundial, J. N. and Kapoor, H. M., *Jour. Pal, Soc. Ind.,* 1973, 17, 50.
4. Gupta, V. J., *Indian Palaeozoic Stratigraphy,* Hindustan Publishing Corporation, Delhi, 1973, p. 207.
5. Ravi Shanker and Ganesan, T. M., *Himalayan Geology,* 1973, 3, 72.
6. Stauffer, C. R., *Jour. Pal.,* 1939, 13, 500.

FIRST RECORD OF THE MICROSPORIDIA *NOSEMA* SP. ON *PLUSIA PEPONIS* F. FROM INDIA

IN India, the occurrence of microsporidia *Nosema* sp. has been reported on Honeybees[1] and on silk worms[2]. But no record of *Nosema* sp. was reported on *Plusia peponis* from India. Recently, a group of *P. peponis* larvae collected from snakegourd around the University Campus, Coimbatore, was found dead. The tissue smears, revealed numerous spores of *Nosema* sp. (Fig. 1) in the fat body. The spores were harvested by differential centrifugation. A heavy spore suspension in distilled water was found to be infective on inoculation to disease free healthy larvae by ingestion. On piercing the infected larvae with a needle, a whitish body fluid oozed out which had spores.

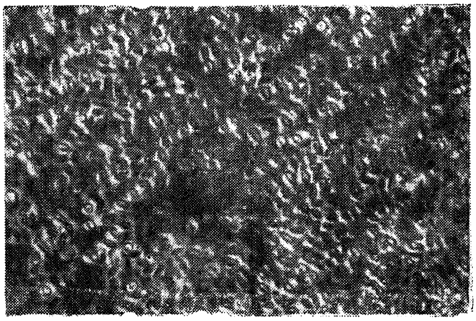

FIG. 1. Spores of *Nosema* sp. in *Plusia peponis* under phase contrast.

This appears to be the first record of the *Nosema* sp. on *P. peponis* from India.

Our thanks are due to Dr. Gerard M. Thomas, Insect Pathology Laboratory, University of California, for his kind help in identifying the pathogen.

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1. Hassanein, M. H., *J. Zool. Soc. Bang.,* 1953, 5, 1.
2. Talukdar, J. W., *Curr. Sci.,* 1961, 30, 28.

HOST CORRELATED VARIATION IN *TRIALEURODES RARA* SINGH AND *BEMISIA TABACI* (GENNADIUS) (ALEYRODIDAE : HOMOPTERA : INSECTA)

THE last larval exuvium or the pupal case in most aleyrodids remaining closely attached to the leaves of their host plants, offer a multitude of diagnostic features aiding in the proper placement of the concerned genera and species. As adult aleyrodids are not always associated with their hosts, the identity of species based on a collection of such pupal cases from their host plants is of considerable significance not only in understanding the host range, but also the impact of different host plants on the degree of intraspecific diversity of the concerned species. Takahashi[1] (1933) established a correlation between the structure of the pupal cases of species with the nature of the leaves of the host plants. The variations in the number and size of the setae were reported in a few species like *Bemisia tabaci* by Husain and Trehan[2] (1933), in *Aleyrodes fragriae* Walker by Deshpande[3] (1933) and in *Asterobemisia caprini* (Koch) by Trehan[4] (1940). Russel[5] (1948) indicated the effect of the physical nature of the leaves on the morphology of the species of *Trialeurodes* and indicated that the genus as a whole is considerably plastic. Takahashi[6] (1955) reported variations in the size of the dorsal setae and pupal cases found among the individuals in the same colonies of *Pealius rubi* Takahashi on many of its hosts. Mound[7] (1967) in his work on host correlated variations in *Bemisia tabaci* established through experimental work, the existence of a number of separable morphological forms, earlier considered as distinct species.

An attempt has been made here to provide a comparative idea of the degree of intraspecific diversity in *Bemisia tabaci* and *Trialaleurodes rara* on different host plants and the variations studied with respect to several characters cited in Tables I and II.

TABLE I

Host correlated variation in certain characters of the pupal case of Bemisia tabaci

Nature of leaves	Siliceous	Hairy		Partially hairy	Glabrous	
Host plants	*Oryza sativa*	*Gauzuma tomentosa*	*Physalis peruviana*	*Gossypium hirsutum*	*Aristalochia bilobosa*	*Lab lab niger* (= *Dolichos lablab*)
Length (mm)	0·83–0·85	0·46–0·47	0·57–0·59	0·57–0·59	0·58–0·64	0·57–0·64
Width (mm)	0·47–0·50	0·30–0·39	0·37–0·40	0·44–0·47	0·44–0·47	0·42–0·48
Elongated dorsal setae	One on cephalic region	Seven	Seven	None	None	None
Dorsal pores and porettes	Present	Absent	Absent	Present	Absent	Absent
Margin	Crenulate irregular	Crenulate but sometimes deeply indented	Crenulate, irregular	Smooth, crenulate at tracheal pores	Crenulate	Smooth or fairly irregular
Thoracic tracheal folds	Present	Present	Present	Present	Present	Present
Median tubercles	Absent	Two	Absent	One or none	Two	Rarely one
Subdorsal abdominal papillae	Absent	Poortly defined	Absent	Poorly defined	Six, but ill-defined	Frequently absent
Subdorsal area	Finely rugose	Narrow and rugose	Narrow and rugose	Smooth or finely rugose	Broad and smooth	Broad and smooth
Caudal ridges	Slightly indicated	Clearly visible	Clearly visible	Poorly defined	Poorly defined	Poorly defined
Vasiform orifice (mm)	0·071–0·094 × 0·052–0·058	0·047–0·055 × 0·038–0·041	0·058–0·061 × 0·041–0·044	0·055–0·058 × 0·041–0·047	0·063–0·072 × 0·044–0·047	0·058–0·063 × 0·047
Lingula tip (mm)	0·033 × 0·016–0·019	0·016–0·019 × 0·014	0·022–0·025 × 0·011–0·014	0·022×0·017	0·019–0·025 × 0·016–0·019	0·022–0·025 × 0·014–0·017
Operculum (mm)	0·033–0·039 × 0·044–0·050	0·022–0·025 × 0·030–0·033	0·025–0·030 × 0·033–0·036	0·027–0·030 × 0·036	0·030–0·033 × 0·033–0·039	0·030 × 0·036–0·047
Caudal furrow (mm)	0·066–0·067	0·036–0·044	0·049–0·052	0·046–0·050	0·041–0·050	0·041–0·044

It may be evident that the pupal cases from *Oryza sativa* are very elongate, bearing a pair of

TABLE II

Host correlated variation in certain characters of pupal case of Trialeurodes rara

Naturc of leaves	Glabrous				Partially hairy	Hairy
Host plants	*Aristolochia labiosa*	*Lal bab niger* (= *Dolichos lab lab*)	*Phyllanthus acidus*	*Ricinus communis*	*Gossypium hirsutum*	*Bauhinia* sp.
Length (mm)	0·05–0·56	0·48–0·64	0·46–0·47	0·53–0·57	0·54–0·55	0·44–0·51
Width (mm)	0·33–0·37	0·33–0·41	0·30–0·31	0·31–0·37	0·38–0·40	0·30–0·32
Number of strong crenulations at thoracic tracheal ending	5–7	8	8	8	8–14	7
Number of strong crenulations at caudal ending	6	8	4	6	6	8
Cephalic setae (mm)	0·017	0·014	0·017	0·014	0·014	0·019–0·022
First abdominal setae (mm)	0·014	0·011	0·017	0·014	0·014	0·019
Eighth abdominal setae (mm)	0·014	0·014	0·014	0·014	0·014	0·011–0·014
Caudal setae (mm)	0·039	0·044	0·041	0·044	0·044	0·044
Vasiform orifice (mm)	0·055 × 0·044–0·050	0·052–0·055 × 0·041–0·050	0·050–0·058 × 0 041–0·044	0·050–0·055 × 0·044–0 050	0·055–0·058 × 0·044	0·047–0·052 × 0·041–0·050
Operculum (mm)	0·027–0·030 × 0·033–0·036	0·025–0·033 × 0·030–0·039	0·028–0·030 × 0·030–0·033	0·030×0·030	0·033×0·036	0·022–0·028 × 0·030–0·036
Number of submarginal papillae	48–54	41–57	52–46	48–50	56–59	30–56
Number of subdorsal papillae	5–8	5–6	5	6	6	10–30

elongate setae on the cephalic region, differing very much in shape from the typical *B. tabaci* on one of its many dicotyledonous hosts. The number of elongate setae is seven in the case of pupal cases on hairy leaves, and none in those taken on glabrous leaves. The data also indicate that in spite of its extremely common occurrence on a variety of hosts plants, *Bemisia tabaci* is unique for its instability of characters as revealed by the striking intraspecific variations. *Trialeurodes rara* in so far as has been investigated also shows this tendency to a remarkable degree, with variations in the number of papillae on submargin and subdorsum as also in the length of the dorsal setae and size of the pupal cases. The ability of these species as well as of others such as *Pealius* and *Dialeurodes* to develop on particular types of leaves, leading to the development of considerable plasticity of the species appears to be a direct reaction to the damage of their immediate environment.

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---

1. Takahashi, R., *Formosa Govt. Res. Inst. Dept. Agr. Rept.*, **60**, 1.
2. Husain, M. A. and Trehan, K. N., *Indian J. Agric. Sci.*, 1933, **3** (4), 701.
3. Deshpande, V. G., *Trans. R. ent. Soc. Lond.*, 1933, **81**, 117.
4. Trehan, K. N., *Ibid.*, 1940, **90**, 575.
5. Russel, L. M., *Misc. Publs. U.S. Dept. Agric.*, 1948, **635**, 85.
6. Takahashi, R., *Mushi*, 1955, **29** (3), 9.
7. Mound, L. A., *Proc. R. ent. Soc. Lond.*, 1967, **38 A**, 171.

REGULATION OF URIC ACID SYNTHESIS DURING AESTIVATION BY GLUTAMINE LEVELS IN *PILA GLOBOSA*

EARLIER reports on aestivation indicated that uric acid synthesis is put up in the early stages of aestivation in *Pila globosa*[1] and consequently glutamine levels[2] and glutamine synthetase activity[3]. The glutamine synthetase was known to increase so as to meet the needs of uric acid synthesis, though the cellular environment is not favourable for the activation of the concerned enzyme[3]. The rate of uric acid synthesis is decreased in the digestive gland and mantle of *Pila globosa* after three months of aestivation[4]. Hence it would be more pertinent to study the levels of the enzyme, glutamine synthetase, after three months of aestivation. The present report concerns the estimation of the enzyme by the method of Iqual and Wu[5] in the digestive gland, mantle and foot of active and of 6 months aestivated *Pila globosa.*

TABLE I

Glutamine synthetase activity in the digestive gland of active and 6 months aestivated Pila globosa

Sample	Activity, μ moles of γ glutamyl hydroxamate formed/gm. wet wt. of tissue/hr. values mean $\pm$ S.D.
Active	$6 \cdot 3 \pm 1 \cdot 8$
Aestivated	$2 \cdot 6 \pm 1 \cdot 2$

Level of significance, p value $< 0 \cdot 001$.

Note.—Each value is the average of 10 separate experiments.

The activity of glutamine synthetase has decreased by 59% in the digestive gland of 6 months aestivated *Pila globosa* (Table I). The other two tissues, *viz.,* mantle and digestive gland had no detectable enzyme activity, both in the active and aestivated snails pointing towards the pivotal role of glutamine levels in limiting the uric acid biosynthesis. Similar function for glutamine synthetase in the control of nitrogen metabolism where the glutamine levels autoregulate their flow either towards urea synthesis or uric acid synthesis had been envisaged in terrestrial gastropods[5]. The decrease of the enzyme in 6 months aestivated *Pila globosa* may be due to the different modes of control on the enzyme. *Pila globosa* was believed to rely on the carbohydrate and fat reserve after 3 months of aestivation. Hence an increase in glutamine synthetase activity after 3 months of aestivation is not warranted. But earlier studies in 3 months aestivated snails showed that the cellular environment which favours adenylylation or deactivation of the enzyme is compensated by active deadenylylation thus increasing the activity of glutamine synthetase during this period. The decrease in the activity of glutamine synthetase after 3 months may be due to the loss of this control by deadenylylating system. But such a conclusion is also not appropriate since the synthesis of each adenylylating complex requires 12 ATP which will be a wasteful dissipation of high energy molecules. It remains to be seen whether the enzyme is reguated at the genetic level in the control of enzyme concentration itself.

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---

1. Srinivasa Reddy, Y., Venkateswara Rao, P. and Swami, K. S., *Indian J. Expt. Biol.,* 1974, 12, 454.
2. Ramana Rao, K. V., *Ph.D. Thesis,* S.V. University, Tirupati, 1974.
3. Srinivasa Reddy, Y. and Swami, K. S., *Curr. Sci.,* 1975, 44 (6), 191.
4. —, *Ph.D. Thesis,* S.V. University, Tirupati, 1974.
5. Iqual, K. and Wu, C., *Enzyme,* 1971, 12, 553.
6. Hochachka, P. W. and Somero, G. N., *Strategies of Biochemical Adaptation,* W. B. Saunders Company, 1973, p. 171.
7. Raghupatirami Reddy, S., *Ph.D. Thesis,* S.V. University, Tirupati, 1965.

EFFECT OF HYPOPHYSECTOMY ON SERUM CHOLESTEROL IN CATFISH, *HETEROPNEUSTES FOSSILIS* BLOCH.

THE pituitary gland seems to be essential for cholesterol utilization, because its surgical removal results in hypercholesterolemia[1-3]. However, it also influences the secretion of other endocrine glands and, therefore, its absence for a long period would result in hormonal imbalance, which, in turn, would affect cholesterol utilization. Since such a study is not reported earlier, the present work was undertaken to find out hypophysectomy effects on serum cholesterol level at intervals of one, two, four and fifty weeks. The results reported in this investigation are based on serum cholesterol level in 21 hypophysectomized and 22 sham-hypophysectomized (controls) animals. The source of fish, their laboratory care and the method of hypophysectomy are described elsewhere[4]. The periods during which the hypophysectomies were performed were September 1972, February 1973 and May 1973. The animals, which were hypophysectomized during May 1973 were sacrified one and two weeks after the operation, whereas those operated during February 1973 and September 1972 were sacrificed four and fifty weeks after the operation. Blood samples were drawn at autopsy from the caudal veins and the cholesterol content was determined colorimetrically according to the method of Abell *et al.*[5]. Statistical analysis was based on Student's '*t*' test.

It is seen from the results (Table I) that there is no significant difference between the serum cholesterol levels in the controls and in the hypophysectomized animals within a period of one week. However, the serum cholesterol level becomes almost double within two (810 ± 37 mg/100 ml) and four (1065 ± 84 mg/100 ml) weeks after hypophysectomy as compared to the cholesterol values in the corresponding controls (410 ± 28 mg and 430 ± 39 mg/100 ml). After one year the serum cholesterol level is depleted in the hypophysectomized animals as compared to that in the other experimental groups, although it is still higher than the corresponding control level. These observations indicate the presence of hypocholesteromic pituitary factor(s) in *Heteropneustes fossilis*. The depletion of cholesterol level in hypophysectomized animals after one year is a significant feature and is probably due to the hormonal imbalance as a result of hypophysectomy which has affected the mobilization and/or utilization of cholesterol in this fish.

TABLE I

Effect of hypophysectomy on serum cholesterol (mg/100 ml) in Heteropneustes fossilis

Duration (weeks)	Controls	Hypophysectomized fish
1	380±40 (6)	420±60 (6)
2	410±28 (5)	*810±37 (5)
4	430±39 (6)	*1065±84 (6)
50	524±52 (5)	*670±47 (4)

* P< 0·001.

Number of animals is given in parenthesis.

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---

1. Friedman, M., Byers, S. O. and Fleck, S. R., *Nature* (*Lond.*), 1970, **228**, 464.
2. Byers, S. O., Friedman, M. and Roseman, R. H., *Ibid.*, 1970, p. 228.
3. Srivastava, A. K. and Pickford, G. E., *Gen. Comp. Endocrinol*, 1972, **19**, 290.
4. Belsare, D. K., Belsare, S. G. and Murthy, P. R. S., *Acta Zoologica.*, 1970, **51**, 219.
5. Abell, L. L., Levy, B. B., Brodie, B. B. and Kendall, F. E., *J. Biol. Chem.*, 1952, **195**, **351**.

THE EFFECT OF HEAT TREATMENT ON THE MOISTURE, VOLATILE MATTER AND CALORIFIC CONTENT OF THE DEPOT FATS OF SOME FRESHWATER FISHES

DESPITE considerable work on the chemistry of fish lipids[1-8], information on the moisture and volatile matter and the relation between volatility and the calorific value of the fats of Indian freshwater fishes seems to be lacking. The present study which is based on the quantitative estimates of moisture, volatile matter and energy content in the depot fats of four species of freshwater teleosts, namely, *Rita rita* (Ham.), *Heteropneustes fossilis* (Bloch.), *Wallagonia attu* (Bloch.) and *Ophicephalus punctatus* (Bloch) is an attempt in this direction.

The methods for the determination of moisture and volatile matter in the fat and that of heat treatment were the same as given in the A.O.A.C.[9]. All values have been calculated on wet weight basis.

The concentrations of the two constituents were found to differ in different species of fishes (Table I). In the teleosts investigated, the moisture and volatile matter were found to be least concentrated in the cat-fish, *Rita rita*, while their con-

TABLE I

Moisture, volatile matter, and loss in calorific content during heat treatment of the depot fats of some freshwater teleosts

Species of fish	Moisture and volatile matter (%)	Loss in calorific content during heat treatment (calories per 100 g fresh depot fat)
Rita rita (Ham.)	4·650	43·245
Heteropneustes fossilis (Bloch.)	7·278	67·685
Wallagonia attu (Bloch.)	8·965	83·374
Ophicephalus punctatus (Bloch.)	13·636	126·814

centrations were highest in the murrel, *Ophicephalus punctatus*. Since drying of lipids during various technological processing has been reported to lead to a substantial loss in the quantity of more volatile fractions, such as short-chain fatty acid methyl esters and some of the shorter-chain fatty acids[10], the loss in the calorific value of fish lipids would depend directly upon the degree to which they are volatile. Thus, the loss in the calorific value of the depot fat of *Ophicephalus punctatus*, with high volatile matter was greater than that in *Rita rita*, with low volatility (Table I).

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---

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1. Lovern, J. A., *J. Sci. Food Agric.*, 1956, 12, 729.
2. —, *Proc. Nutrition Soc. Engl. Scot.*, 1958, 17, 161.
3. Olley, J., *Biochem. J.*, 1961, 81, 29.
4. Tiwari, R. D. and Srivastava, K. C., *Proc. Nat. Acad. Sci. India*, 1962, 32, 111.
5. — and —, *Ibid.*, 1963, 33, 11.
6. Love, R. M., *The Chemical Biology of Fishes*, Academic Press, London and New York, 1970.
7. Worthington, R. E., Boggess, T. S. and Heaton, E. K., *J. Fish. Res. Bd. Canada*, 1972, 29, 113.
8. Tinsley, I. J., Krueger, H. M. and Saddler, J. V., *Ibid.*, 1973, 30, 1661.
9. A.O.A.C., *Official Methods of Analysis of the Association of Official Agricultural Chemists*, 9th Edn., 1960.
10. Colowick, S. P. and Kaplan, N. O., *Methods in Enzymology*, Academic Press, New York and London, Vol. XIV, 1969.

DIURNAL RHYTHMS IN MITOSIS OF CELLS OF THE WHEAT ROOT TIP MERISTEM

PREVIOUS studies have shown that diurnal rhythms occur in mitosis in the root tip of several plant materials[3,4,6]. Of these the onion showed a four-peaked diurnal rhythm in mitotic activity in root tips grown in darkness at constant temperature[1,2]. This in fact, is contrary to the view held by several other workers including Lewis and Wolff *et al.*[7] who observed two peaks of cell division in roots of onion bulbs. However, no such information is available on diurnal fluctuations in mitosis in cells of wheat root tip meristem. This is a brief report on the rates of mitosis of cells in the wheat root tip meristems under variable conditions of illumination at a fixed temperature.

Seeds of wheat variety C 591 were sown at 1 hour intervals for 24 hours in 52 mm petridishes on Whatman 40 filter-paper moistened with 5 ml of distilled water and were kept in the incubator in complete darkness at 20 ± 1° C. Same procedure was followed when seeds were grown under continuous illumination provided by 300 watt incandescent Lamp at 20° ± 1° C. Twenty seeds were germinated in such petridish. After 72 hours, 3 mm long roots were pretreated with a saturated solution of α-bromo napthalene for 1 hour at 20 ± 1° C, followed by distilled water. They were fixed in 1 : 3 aceto-alcohol. Roots were hydrolysed in 1N HCl for 15 min. at 60° C and stained in Feulgen reagent. Three roots were examined in each collection period. Root tips of approximately 0·5 mm length were cut and squashed in 1% aceto-carmine and 500 cells from the apical meristem were scored under oil immersion. Cells exhibiting well defined prophases on, through to complete formation of the cell plate in telophase were observed.

The study of frequency of the dividing cells in mitosis in wheat root tip meristem showed a regular wave of cell division during 24 hours when wheat seeds were germinated in darkness. The frequency in mitotic cell division gradually increased starting from 0 hour and reached maximum at 12 noon. Thereafter, there was a gradual decrease in mitotic activity and it became minimum towards 0 hour (Fig. 1 *b*). In contrast, constant illumination abo-

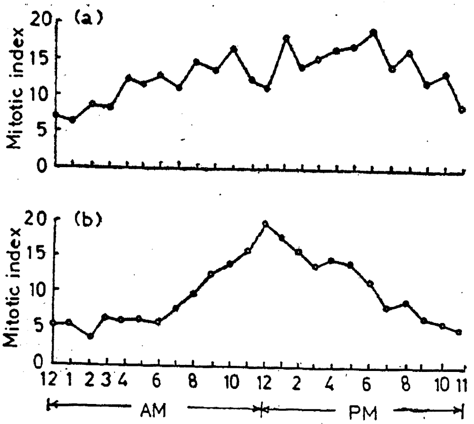

FIG. 1. Mitotic indices in wheat root tip meristem. *a*, Under constant illumination cell division was extremely variable with many irregular peaks and no perceptible mitotic rhythm; *b*, Under constant darkness a single maximum peak in mitotic activity at noon, 12 O' clock.

lished mitotic rhythm and cell division was extremely variable with many irregular peaks; no clear mitotic rhythm was observed (Fig. 1 *a*).

The rate of cell division in wheat root tip meristem followed a diurnal variation when seeds were grown in complete darkness and showed one-peaked diurnal rhythm in mitotic activity at noon. Since soaking period was 72 hours in each case it can be suggested that maximum peak in mitotic cell division was not dependent on the time of imbibation. This is not in conformity with that of Bishop and Klein[1] who observed that mitotic rhythm in onion root tip meristem was dependent upon the time of imbibition and during 24 hours there were four maxima and four minima in mitotic activity. They however, reported complete breakdown in mitotic rhythm in cells of onion root tip meristem following continuous white light illumination. The nature of association between rhythm pattern and time of the day needs further study.

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---

1. Bishop, R. C. and Klein, R. M., *Can. J. Genet. Cytol.*, 1971, 13, 597.
2. —, *Ibid.*, 1973, 15, 667.
3. Friesner, R. C., *Am. J. Botany*, 1920, 7, 380.
4. Jenson, W. A. and Kavalijian, L. G., *Ibid.*, 1958, 45, 365.
5. Kellicot, W. E., *Bull. Torrey Bot. Club*, 1904, 31, 529.
6. Lewis, A. C., *Bot. Gaz.*, 1901, 32, 423.
7. Wolff, E. G., Fiver, D. M. and Klein, R. M., *Bull. Torrey Bot. Club*, 1967, 94, 441.

CHANGES IN AMINO ACIDS AND AMIDES CONTENT IN JOWAR LEAVES INFECTED BY *HELMINTHOSPORIUM TURCICUM*

Helminthosporium turcicum Pass. causes severe leaf blight of jowar (*Sorghum vulgare* Pers.). Decrease[1] or increase[2] in amino acids and amides in infected leaves has been reported in different diseases. But a strange phenomenon was observed in jowar leaves infected with *H. turcicum*. In this case amino acids decreased in young plants, but increased in old plants due to infection. The details are presented in this paper.

Ten- and sixty-day old jowar plants (cultivar Co. 18), were inoculated with equal spore concentration (10^6/ml) of the virulent isolate of the pathogen. After seven days, when the symptoms had fully developed, the lower three leaves were pooled and analysed for their amino acids and amides by the method described by Block *et al.*[3] and for their protein[4]. Two independent experiments were conducted and the data presented in Table I is the average of these two experiments. The data were also statistically analysed.

All the amino acids and amides, excepting asparagine and methionine decreased due to infection significantly in the 10-day old plants. Excepting glutamic acid and glutamine, all the amino acids increased markedly due to infection in the 60-day old plants. There was appreciable amount of reduction in glutamine and glutamic acid contents and negligible amount of reduction in asparagine content due to infection in the latter plants (Table I). When protein nitrogen content was analysed, its content in healthy and infected leaves was 0·31 and 0·39% respectively in 10-day old plants and

TABLE I

Amino acids, amides content of healthy and H. turcicum *infected sorghum leaves*

Amino acids/amides in µg/g of fresh leaves	10-day old plants			60-day old plants		
	Uninoculated	Inoculated	C.D.	Uninoculated	Inoculated	C.D.
Histidine	0	0	..	0	49·0	..
Asparagine	5·4	5·4	N.S.	4·4	4·0	0·2
Aspartic acid	20·5	11·5	0·7	18·3	28·5	0·6
Glutamic acid	13·4	3·1	1·1	6·8	3·8	0·7
Glutamine	296·0	172·0	11·1	108·0	30·4	9·5
Threonine	4·0	0·3	1·1	0·5	2·4	0·4
Alanine	2·1	0·5	0·4	0·5	9·0	1·3
Methionine	2·3	2·1	N.S.	3·0	17·6	0·1
Leucine/isoleucine	2·2	1·3	0·4	9·6	10·8	0·9
Total amino acids and amides	345·9	196·2	15·2	151·1	155·1	3·8

C.D. = Critical difference. N.S. = Not significant. — = Not analysed.

0·24 and 0·02% respectively in 60–day old plants.

The strange phenomenon in which the same amino acids which decreased due to infection at young age of the plants, increased at their old age is not known. But it may be seen that young leaves had more free amino acids than the old leaves. The fungus which depends upon the host amino acids for its growth[5] would have reduced the amino acids content in the young age of the plants. At the old age, the free amino acids present would not have been sufficient for the fungus and the pathogen which is having the capacity to produce proteolytic enzymes[6] would have degraded the host protein into amino acids. Reduction in protein content of jowar has been observed only in infected leaves of 60–day old plants. Hence the protein amino acids which are surplus over the pathogen's requirement would have accumulated in the infected leaves. However, this phenomenon needs further intensive study. Both glutamic acids and glutamine contents reduced in both young and old leaves due to infection and it has been already observed that these particular amino acid and amide induced the maximum growth of the pathogen *in vitro*[6].

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---

1. Shishiyama, J., Egawa, H., Mayama, S. and Akal, S., *Mem. Coll. Agric. Kyoto. Univ.*, 1969, **95**, 7.
2. Singh, D. and Smalley, E. B., *Phytopathology*, 1969, **59**, 891.
3. Block, R. J., Durrum, E. L. and Zweig, G., *A Manual of Paper Chromatography and Paper Electrophoresis*, Academic Press, Inc., New York, 1955, p. 710.
4. Vidhyasekaran, P. and Kandaswamy, D., *Phytopath. Z.*, 1971, **72**, 11.
5. Ohata, K. and Kozala, T., *Ann. Phytopath. Soc. Japan*, 1962, **27**, 73.
6. Arjunan, G., *Studies on the Helminthosporiose Disease of Sorghum*, M.Sc. (Ag.), Thesis, Madras, Univ., 1970, p. 133.

STUDIES ON THE RELATIONSHIP BETWEEN *MELOIDOGYNE INCOGNITA* AND GALLING BEHAVIOUR OF *CICER ARIETINUM* ROOTS

THE polyphagous nature of root-knot nematodes led various workers in the past to study the infestation process with different views. The root-knot infection is brought about by second stage larvae, causing hyperplasia and hypertrophy which result in galls or knots on the infected roots. Consequently, many morphological, anatomical and physio-biochemical abnormalities occur in the host. Hence to understand the host-parasite relationship the attempts were made to study the galling behaviour of the roots of Bengal gram (*Cicer arietinum* L.) in presence of root-knot nematode, *Meloidogyne incognita* (Kofoid and White, 1919) Chitwood, 1949.

Seedlings of Bengal gram variety T-13 were raised singly in 4 cm glass tubes containing acid leached, thoroughly washed, steam sterlized and moistened sand (particle size ranging between 125 μ and 500 μ). The seedlings were supplied daily with Long Ashton nutrient solution[3]. Demineralized water was also supplied whenever necessary. The tubes were kept in the glasshouse at 29° ± 1° C.

One week old seedlings were transplanted from glass tubes in small gelatin capsules, each containing 1000 second stage larvae of *Meloidogyne inco*gnita, obtained from the pure culture. These capsules were then burried in the same glass tubes, and the seedlings were allowed to grow in the glasshouse conditions, for 3, 6, 12 and 24 hours after inoculation. The experiment was quintuplicated.

The plants were then uprooted, and the roots were washed thoroughly to remove the adhering soil particles, fixed in FAA for 24 hours and then examined under the stereoscopic microscope. To determine the 'presence of nematode larvae, the roots were stained with acid-fuchsin lactophenol[8].

The present study revealed that the nematode larvae had gained their entry within 3 hours but they could not produce any visible symptoms on the roots of *Cicer*. The rapidity of single and multiple penetration of cowpea and pineapple roots by root-knot nematode larvae was reported in less than 6 hours[2]. Furthermore, *Heterodera trifolii* was observed in clover roots within 15 minutes after coming in contact with the root surface[6].

In a period of 6 hours, slight swellings occurred near the root-tips which eventually transformed into 'snake-head-like' swellings within 12 hours and later became distinct knots within 24 hours of inoculation. Knot formations were confined only to the root tips and were absent from other portions during this period, although the larval presence was noted in other root zones. Similar observations of nematode penetration at the terminals as well as farther up the roots were reported earlier[1,5,6].

In the pesent findings, the swelling of roots tips, as initial invasion sites suggests that the nematode larvae preferred soft apical meristematic zone of the roots rather than the hard mature parts. The preferential penetration of apical meristematic zone of root tissue was also reported by other workers[1,4,7]. The absenee of knots from other parts of the root system appears to be due to short observation periods in the present study.

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---

1. Christie, J. R., *Phytopathology*, 1936, **26**, 1.
2. Godfrey, G. H. and Oliveira, J., *Ibid.*, 1932, **22**, 325.
3. Hewitt, E. J., *Sand and Water Culture Methods Used in Study of Plant Nutrition*, Commow. Agri. Bur. Farnham, Bucks, England, 1966, p. 547.
4. Jamal, A., *Proc. 62nd Sess. Ind. Sci. Congr.*, New Delhi, 1975, **3**, 80.
5. Krusberg, L. R. and Nielsen, L. W., *Phytopathology*, 1958, **48**, 30.
6. Mankau, R. and Linford, M. B., *Ill. Agri. Exp. Sta. Bull.*, 1960, p. 50.
7. Nemec, B., *Das Problem der Befruchtungsvorgange und andere zytologische Fragen* VI. *Vielkernige Riensenzellen in Heterodera Gallen*, Gebruder Borntrager, Berlin, 1910, p. 151.
8. Southey, J. F., *Laboratory Methods for Work with Plant and Soil Nematodes*, Her Majesty's Stationary Office, London, 1970, p. 55.

EFFECT OF WETTING AND DRYING OF THE SOIL ON THE LESION LENGTH PRODUCED BY *XANTHOMONAS ORYZAE*

IT was reported in Japan[1,3], that under alternative irrigation and drainage, the intensity of bacterial blight disease was greater than under continuous irrigation. However, information is lacking on the effect of alternate wetting and drying of the soil before and after the entry of the pathogen into the host. Experiments were therefore conducted in pots with 60 days old plants of *Taichung* (*Native*)[1]. Pin-prick[2] method of inoculation was employed using an inoculum concentration of 1·0 OD.

One set of the test plants was kept submerged to a depth of 5–6 cm and the water was withdrawn before and after inoculation, at intervals of 1, 2, 4 and 8 days. In another set, the plants were maintained under saturation and given submergence treatment for 1, 2, 4 and 8 days before and after inoculation. The details are given in Table I. There was a set of control pots for continuous wetting and continuous drying. There were four pots for each treatment with two plants in each. The lesion length was measured 14 days after inoculation.

The data on the lesion (Table I) showed that continuous drying treatment resulted in significantly less lesion length (4·7 cm) than that of the continuous wetting treatment (9·0 cm).

TABLE 1

Effect of wetting and drying of the soil at different intervals before and after inoculation on the length of the lesion

Treatment	Lesion length (cm)	Treatment	Lesion length (cm)
Continuous wetting of the soil	9·0	Continuous drying of the soil	4·7
Drying the soil before inoculation		Wetting the soil before inoculation	
1 day	6·8	1 day	7·3
2 days	5·3	2 days	7·7
4 days	4·4	4 days	7·8
8 days	4·1	8 days	8·3
Drying the soil after inoculation		Wetting the soil after inoculation	
1 day	3·2	1 day	9·4
2 days	3·9	2 days	8·4
4 days	3·4	4 days	7·2
8 days	4·3	8 days	7·3

Grand Mean 6·27, C.D. at 5%= 1·3 and at 1%=1·8.

When the soil was dried from continuous wetting condition before inoculation, there was a gradual decrease in the lesion length from 1–8 days treatment but there was a drastic reduction in lesion length if the drying was done after inoculation. Drying the soil 4 and 8 days before inoculation and 1–8 days after inoculation gave similar results in respect of lesion length.

When the soil was wetted from continuous saturated or "dry" condition, there was a significant increase in the lesion length as compared to the control (continuous drying of the soil). But the treatment of wetting for 1–8 days before inoculation did not differ significantly from one other.

Wetting the soil a day after inoculation resulted in the longest lesion length of 9·4 cm which was at par with the continuous wetting control (9·0 cm). However, there was a slight reduction in lesion length when the soil was wetted at 4 and 8 days after inoculation as compared to 1 and 2 days.

It might be concluded that submersion of soil was associated with higher infection and drying the opposite effect. Alternative irrigation and drainage could therefore play significant role in the overall disease build-up. The current results also corroborate the observation made by Yamatoki[9] who reported that the disease was severe in shallow field with much moisture fluctuation than in a field with constant soil moisture. Contrary to the report of Kondo and Harada[7], the lesion spread was more in plants grown under continuous wetting condition, thus confirming the observations made by other workers[5,6,8].

The vascular system serves as a pathway for the movement of water and essential metabolites. It was pointed out by Goodman *et al.*[4], that factors which alter the availability of water to the plant cell or impair the movement of water from one cell to another may cause profound alterations in permeability, photosynthesis, cell-wall metabolism, carbohydrate metabolism and other functions. Therefore, a satisfactory explanation for the variation in the lesion length in different treatment has yet to be proposed.

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1. *All Prefectures in Hokuriku.*, Report on bacterial leaf blight (Mimeo.), 1959.
2. Devadath, S. and Padmanabhan, S. Y., *Plant Dis. Reptr.*, 1969, 53 (2), 145.
3. Fukuoko, Agr. Exp. Sta., *Fukuoka Agr. Expt. Sta. Spec. Rept.*, 1920, p. 1.
4. Goodman, R. N., Király, Z. and Zaitlin, M., *The Biochemistry and Physiology of Infectious Plant Disease*, D. Van Nostrand Company, Inc., U.S.A., 1967.
5. Kojima, K., *B. Aichi. Agr. Expt. Sta.*, 1954, 9, 151.
6. —, Iwase, S. and Iki, K., *Ibid.*, 1954, 9, 157.
7. Kondo, G. and Harada, T., *Agr. Hort.*, 1951, 26 (5), 533.
8. Nishida, T., *Noji. Zappo.*, 1909, 127, 68.
9. Yamatoki, T., *Byochugai Zassi.*, 1920, 7, 689.

PRODUCTIVE EFFICIENCY OF SEMI-DWARF CULTURES DERIVED FROM *JAPONICA* AND *INDICA* RICE HYBRIDS

THE growth pattern and yield potential of four recently evolved early (110–115 days) *japonica* × *indica* (*J* × *I*) cultures[1] were compared with that of six high yielding cultures derived from Dee-geo-Woo-gen (DgWg) parentage.

The cultures were grown during *kharif* 1974 under 50 and 100 kg N/ha. Nitrogen was applied in 3 splits ($\frac{1}{2}$ at planting + $\frac{1}{4}$ at mid-tillering + $\frac{1}{4}$ at primordial initiation); P_2O_5 and K_2O at the rate of 50 kg each/ha were applied at planting. The experiment was conducted in a split plot design with N rates in main plot and cultures in sub-plot. Three replications were maintained with a plot size of 2 m × 6 m and spacing of 20 cm × 15 cm. Observations were recorded on tillering, dry matter production and LAI. Yield and yield components were assessed at harvest.

The data are presented in Table I. The *J* × *I* cultures gave invariably higher yield than the cultures derived from DgWg gene source. Among the *J* × *I* cultures, *J* × *K*–34–278 (*Jikkoku* × *Kolamba*–540) gave the highest yield (4·84 t/ha) even at 50 kg N rate. The differences between the two N rates (50 and 100 kg N/ha) was not significant indicating that 50 kg N/ha was optimum. In view of their short duration, the yield obtained may be considered satisfactory.

The grain yields in *J* × *I* cultures were associated with yield attributes such as high panicle number/m², moderate grain number/panicle and high grain

TABLE I

Grain yield and associated characters of J × I and other cultures during kharif *reason at 50 kg N/ha.*

	Cultures	Yield t/ha	Panicles/ m²	Grain No./ Pan	1000 g wt. (g)	At flowering			HI
						DM-t/ha	LAI	SLW-mg/dm²	
A	*J × I cultures*								
	J × *K*-34–278	4·84	473	45	19·6	4·85	3·93	328	60
	J × *S*-62–102	4·53	509	46	19·2	4·52	3·81	342	67
	J × *CR*-1014–162	4·41	450	40	19·9	4·52	4·12	320	59
	H × *Z*-17–511	4·37	507	42	19·4	4·29	3·65	344	56
	Mean (A)	4·54	486	44	19·5	4·55	3·85	334	61
B	*Other cultures/Varieties* (DgWg source)								
	CR-141–4004–192	4·32	411	39	23·1	3·84	3·13	350	62
	CR-110–174	4·25	377	47	20·5	3·79	2·50	387	57
	CR-126 42–1	4·25	479	38	21·2	3·90	2·54	405	61
	Ratna	4·11	433	43	20·6	4·01	3·03	392	47
	Pusa-2–21	4·19	441	41	21·7	3·91	2·64	402	55
	CR-44–36	3·80	417	39	21·2	4·29	3·27	366	47
	Mean (B)	4·15	426	41	21·4	3·96	2·85	384	55
	(A)/(B) × 100	110	114	108	91	115	136	85	111
	C.D. (0·05)	0·43	44	2·7	0·40	0·41	0·37	26	4

number/m^2 and growth attributes like high dry matter and LAI at flowering, thin leaves, late senescence of leaves and high harvest index when compared with other cultures confirming that dry matter and LAI at flowering were significantly associated with yield during *kharif*[2]. These cultures also possessed fine grains with good cooking quality and showed some tolerance to major pests and diseases even under high nitrogen rate (100 kg N). $J \times I$ hybrids appear to be better adapted to *kharif* conditions when compared with cultures one of the parents of which DgWg was the gene donor[1] (Fig. 1).

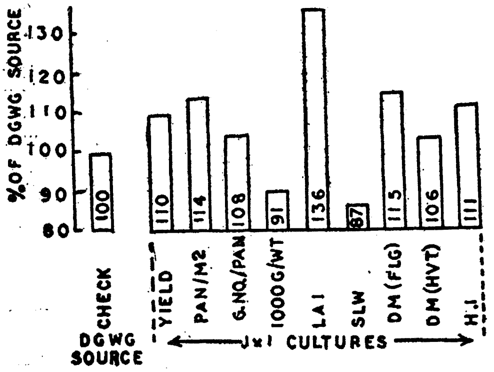

FIG. 1. Mean grain yield and other attributes of $J \times I$ cultures as percentage of check (DgWg source) varieties/cultures.

Thanks are due to Sri M. J. Balakrishna Rao, Geneticist and Botanist, for the supply of $J \times I$ cultures and helpful suggestions.

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K. S. MURTY.

---

1. Rao, M. J. B. K. and Nagaraju, M., *IRC Newsletter*, 1974, 23 (1), 41.
2. Murty, K. S., Sahu, G. and Nayak, S. K., *Indian J. Genet.*, 1973, 34 A, 995.

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Bangalore-6,
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SHORT SCIENTIFIC NOTES

Cucurbita pepo DC, A New Host to *Phoma herbarum* Westend

A severe leaf spot infection of *Cucurbita pepo* DC was observed near Guntur, Andhra Pradesh, during the present rainy season (September–October, 1975). This disease, a new record for India, is reported here.

The leaf spot disease was manifested by the presence of large, dark brown necrotic lesions along the margins of the leaves and oblong to oval, necrotic spots (0·8 cm × 0·5 cm) on the lamina which gradually increased in size covering the entire lamina. The disease spreads from basal to upper leaves. Isolations from several necrotic lesions on Czapek–Dox agar medium yielded a pycnidial fungus. The pathogenecity of the fungus isolated from the lesions was proved by spraying the spore suspension on detached, healthy twigs and covering with moist polyethelene bags for three days. Typical symptoms appeared in 3–5 days after inoculation. Reisolations yielded the original fungus.

Mycelium brown-black with rough surface, hypha dark brown, septate, pycnidia dark brown, numerous, globose to subglobose, pycnidiospores hyaline, single celled, oval to elliptical and measure 5–10 μ × 2–4 μ. The fungus was identified as *Phoma herbarum* Westend.

One of us (CSKV) is thankful to CSIR, New Delhi, for awarding Junior Research Fellowship.

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Guntur 522 005 (AP), *October* 27, 1975.

Trichoderma harzianum—A New Record from India

During a survey of diseases of sunflower in February 1975, root-rot (*Sclerotium rolfsii*) affected plants were collected at Regional Research Station, Mandya, Karnataka. A closer examination of the affected plant parts revealed greenish moldy growth at the collar region. Isolations from these portions yielded a species of *Trichoderma*.

Colonies on potato dextrose agar spreading, floccose, white at first, becoming green in five to six days, reverse side of the colony colourless, vegetative hyphae septate, hyaline, branching, chlamydospores globose, hyaline, intercalary. Philades ampuliform, bearing terminal heads composed of subglobose phialospores held in a mucous aggregate. The fungus was identified as *Trichoderma harzianum* Rifai, by Dr. R. A. Samson of C.B.S., Netherlands. The measurements of the present isolate are within the limits of *T. harzianum* described by Rifai[1].

As the fungus was consistently associated with *S. rolfsii* the nature of its relation with the latter was studied on PDA. Both the fungi grew without any apparent antagonism to each other. However, in due course, *T. harzianum* over-grew *S. rolfsii* parasitizing the latter. Attempts to reisolate *S. rolfsii* from such plates were not successful. *T. harzianum* has been employed successfully to control root-rot of groundnut, tomato and lupine[2]. There is no report of this fungus from India and this constitutes a new record from India.

Grateful thanks are due to Dr. R. A. Samson, Centraalbureau Voor Schimmelcultures, Netherlands, for identifying the fungus and to Dr. H. C. Govindu, for facilities.

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Bangalore 560 024, *November* 25, 1975. P. C. Hiremath.

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1. Rifai, M. A., "A revision of the genus *Trichoderma*," *Commonwealth, Mycol. Inst., Mycol. Paper No. 116*, 1969, p. 56.
2. Wells, H. D., Bell, D. K. and Jaworski, L. A., "Efficacy of *Trichoderma harzianum* as a biocontrol for *Sclerotium rolfsii*," *Phytopathology*, 1972, 62, 442.

On the Occurrence of a Rotifer *Asplanchnella sieboldii* (Leydig) *urawaensis* (Sudzuki) in Indian Waters

In the course of studies on rotifers in July, 1975, we have come across a species of the genus *Asplanchna* from the plankton collections of Allipuram Tank, Visakhapatnam, Andhra Pradesh. At the time of collection the temperature was 27·5° C, pH 8·5, dissolved oxygen 6·8 ppm and turbidity 6·5 ppm.

The species described here has a horse-shoe shaped vitellarium and conforms very well with *Asplanchnella sieboldii* (Leydig) *urawaensis* (Sudzuki) reported earlier from Japan[1]. So far 3 species, *Asplanchna priodonta* (Gosse), *A. brightwelli* (Gosse) and *A. intermedia* (Hudson) are described from the Indian water[2,3]. The present species *Asplanchnella sieboldii* var. *urawaensis* is being reported for the first time from India. It has a transparent, soft sock-formed body, which is changeable in shape as the internal and the external organs move. There are twenty-eight nuclei in the vitellarium. The gastric glands round, no intestine, large body cavity and trophi incudate.

The forms measure a total length of 645–680 μm and a maximum breadth of 375–395 μm.

We are grateful to Prof. K. Hanumantha Rao, and one of us (RKR) thanks the C.S.I.R., for awarding a Junior Research Fellowship.

Department of Zoology, R. KAMESWARA RAO.
Andhra University, P. CHANDRA MOHAN.
Waltair, *October* 27, 1975.

---

1. Sudzuki, Minoru, *Hydrobiologia*, 1964, 23, 75.
2. Edmondson, W. T. and Hutchinson, G. E., *Mem. Conn. Acad. Arts. Sci.*, 1934, 10, 158.
3. Arora, H. C., *J. Zool. Soc. India*, 1963, 14, 40.

Sclerotial Wilt of *Oscimum canum* Sims. A New Record from India

Oscimum canum Sims. (Ban Tulasi) grows wild in Gorakhpur (U.P.), locality. Leaves and seeds of this plant are used medicinally. During August, 1975, a severe sclerotical wilt of the plants was observed in this region when high temperatures prevailed after heavy rains. The infection started from the collar region of the plants. In advanced stage of the disease the whole main stem was covered with white mycelial growth bearing sclerotial aggregations. Lastly, the infection resulted into complete wilting.

The organism responsible for the wilt was isolated and studied on potato dextrose agar medium. On the basis of typical morphological features and cultural characteristics, the organism was identified as *Sclerotium rolfsii* Sacc. [*Corticium rolfsii* (Sacc.)] Curz. The pathogenecity of the organism was established by inoculating the plants under natural conditions. Symptoms appeared and manifested similar to those observed in naturally infected plants. Reisolation yielded the same organism.

Sclerotium rolfsii has elicited good deal of attention from the pathologists as it causes collar and root rot of several economically important plants in this country[1-6]. *Oscimum canum*, however, constitutes a new host record for this fungus from India. The specimen has been maintained as GPU Herb. No. 331 in the Departmental Herbarium of the University.

Authors thanks are due to Dr. K. S. Bhargava, Senior Professor of Botany, for facilities.

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September 13, 1975.

---

1. Kamal and Singh, S., *Curr. Sci.* (accepted), 1975.
2. —, and —, *Indian Phytopath.* (accepted), 1975.
3. Butler, E. J. and Bisby, G. R., *The Fungi of India* (1952) (Revised by R. S. Vasudeva), ICAR, New Delhi, 1960, p. 512.
4. Ramakrishnan, K. and Subramanian, C. V., "List of Indian Fungi (1952–1962)," *J. Madras Univ.*, 1956, 26 B, 327.
5. Tandon, R. N. and Chandra, S., *Supplement to the list of Indian Fungi* (1957–1962), Univ. Allahabad Studies, Allahabad, 1963, p. 246.
6. Mukerji, K. C. and Junija, R. C., *Fungi of India, Supplement to the List of Indian Fungi* (1962–1972), Emkay Publication, Delhi, 1975, p. 224.

New Record of *Bacillus cereus* and *Streptococcus* sp. on the Pink Bollworm of Cotton, *Pectinophora gossypiella* (S.)

In India, *Bacillus* Fr. and Fr. has been reported to occur on *Papilio demoleus*[1] and on seven lepidopteran insects[2]. Recently Thontadarya[3] reported it on *Earias vitella*. Though various species of *Streptococci* have been found to cause disease on different insects[4,5], no record of *Streptococcus* sp. and *Bacillus cereus* has appeared on *Pectinophora gossypiella* in India. While examining the bollworm infested cotton bolls from the fields around Coimbatore, the authors noticed some of the dead larvae inside the bolls and were found infected by mixed bacteria. The pathogens were identified as *Bacillus cereus* and *Streptococci* sp. The pinkish white infected body had putrified fluid inside. Mostly the late stage larvae were susceptible to infection.

This appears to be the first record of *B. cereus* and *Streptococci* sp. on *P. gossypiella*.

Our thanks are due to Dr. Gerard, M. Thomas, of the University of California, for identifying the pathogens.

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December 20, 1975.

---

1. Rangaswami, G. and Ramamoorthi, K., *Indian J. Microbiol.*, 1963, 3, 51.
2. —, —, and Oblisami, G., "Studies on Microbiology and Pathology of Insect Pests of Crop Plants. USDA-PL. 480, Research Project," *Final Rep. Uni. Agric. Sci. Hebbal*, Bangalore, 94.
3. Thontadarya, T. S., Holi Hosur, S. N. and Hiremath, I. G., *Curr. Sci.*, 1975, 44, 488.
4. Steinhaus, E. A., *Principles of Insect Pathology*, McGraw-Hill Book Co., Inc., New York, 1949, p. 757.
5. Battu, G. S., Bindra, O. S. and Rangarajan, M., *Indian J. Ent.*, 1971, 33, 317.

Estimation of the Population of *Xanthomonas oryzae* Introduced into Natural Eco-systems

The chief drawback in studying the population dynamics in natural eco-system is the absence of standard procedures to measure accurately the populations of the bacterium when introduced into a heterogeneous microbial community. Work was initiated[1], therefore to incorporate antibiotic resistance in the bacterium to study the population dynamics of the bacterium. Studies carried out elsewhere[2], independently on the role of seed transmission by employing similar technique proved to be successful.

A streptomycin resistant mutant was isolated from isolate 'P' of *X. oryzae* by streaking on Silva and Buddenhagen (SB)[3], medium supplemented with 500 ppm streptomycin sulphate and the colonies which developed were re-streaked serially on plates containing graded concentrations of the antibiotic. At different levels of streptomycin resistance, the bacterial suspension was added to the soil and checked for *X. oryzae* growth on the respective concentration of streptomycin supplemented medium. At 25,000 ppm it was found that all soil bacterial flora could be effectively checked allowing only the growth of streptomycin resistant *X. oryzae*. But some colonies of *Aspergillus* and *Penicillium* spp. were found growing faster overtaking the growth of *X. oryzae*. Mycostatin and actidione were found to be the effective fungicides in suppressing the soil fungal flora at 100 ppm.

The virulence of the bacterium was maintained by inoculating to rice plants and re-isolating on streptomycin and mycostatin incorporated medium thrice.

Ten ml of bacterial suspension of 10^9 cells/ml was added to 100 g of field soil and mixed thoroughly, to which 90 ml of tap water was added, shaken thoroughly and the suspension was allowed to settle down. Serial dilutions of this suspension were made. Petri plates of 10 cm size were poured with 25,000 ppm streptomycin and 100 ppm mycostatin supplemented SB medium and allowed to harden. Three ml of the SB medium supplemented with 25,000 ppm of streptomycin and 100 ppm mycostatin mixed with 1·0 ml of the serial dilution of the soil suspension was poured as the top layer and was allowed to solidify. The plates poured without streptomycin and mycostatin served as control. Four replicate plates were maintained for each treatment and were incubated at 28° C for 6 days and observed for the appearance of colonies of *X. oryzae*.

In control plates no *X. oryzae* could be recovered because of the fast growing saprophytic soil microflora. In the treatment with both 25,000 ppm streptomycin and 100 ppm mycostatin, all the soil bacterial and fungal flora could be suppressed enabling the colonies of *X. oryzae* to grow.

This technique can be used effectively in studying the ecolony of the bacterium, *viz.*, viability in soil, plant debris, etc., without interference from faster growing soil microflora though this technique is limited to streptomycin resistant mutants of *X. oryzae*.

we are thankful to Dr. S. Y. Padmanabhtn, Director, for his sustained encouragement durng this work. We wish to record our thanks to Dr. H. E. Kauffman, Plant Pathologist, International Rice Research Institute, Philippines, for his generous gift of mycostatin.

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1. Central Rice Research Institute, *Annual Report*, 1971, p. 142.
2. Hsieh, S. P. Y., Buddenhagen, I. W. and Kauffman, H. E., *Phytopathology*, 1974, 64, 273.
3. International Rice Research Institute, *Annual Report*, 1971, p. 108.

REVIEWS AND NOTICES OF BOOKS

Interfacial Electrochemistry—An Experimental Approach. By E. Gileadi, E. Kirowa-Eisner, and J. Penciner. (Addison-Wesley Pub. Co., Inc., Reading, Mass. 01867), 1975. Pp. xviii + 525. Price \$ 19.50 (Cloth) ; \$ 13.50 (Paper).

The book *Interfacial Electrochemistry—An Experimental Approach,* by E. Gileadi, E. Kirowa-Eisner and J. Penciner, is mainly devoted to a description of some typical experiments in double layer phenomena and electrode kinetics.

In the first part of the book, some basic concepts are introduced pertaining to the structure of the electrical double layer with and without specific adsorption, electrocapillarity and electrode kinetics with and without adsorption of reaction intermediates under activation as well as diffusion control. A short account of some applications in the fields of corrosion, batteries and electrodeposition are also given.

In the second part of the book, the reader is introduced to the operational amplifier approach to electrochemical instrumentation. This is followed by a detailed description of several experiments designed to illustrate the concepts covered in the first part of the book.

The usefulness of the book is somewhat decreased by the fact that the equipment for virtually all the experiments described in the book are assembled on the basis of operational amplifiers, potentiometric recorders, current-to-voltage converters, etc. Except for experiments requiring a potentiostat or for transient analysis, other experiments described in the book could very well be carried out equally well by less sophisticated and more readily available components such as potentiometers, galvanometers and batteries. Moreover, experimental set-up arranged from such individual circuit components tends to provide the student with a better comprehension of the working of the equipment than when using an integrated instrument bloc. Hence, even assuming an easy availability of the instruments described in the book, electrochemical experiments based on simpler electrical circuit components could have been included to illustrate the main ideas.

There are some mistakes in the exposition of the basic concepts in the book. For example, Z should be only its numerical value and a plus or minus sign attached to the right hand side of equation II, 6. With the convention adopted for the current and overpotential (eq. II, 50), the negative sign on the right hand side of eq. II, 146 is wrong. The explanation of *pseudocapacity* (p. 87) is ambiguous in that one does not directly observe in an experiment the leaky nature of the capacitor ; as very different from an electrical capacitor. The pseudocapacitance is so called because of the observed strong *frequency dependence* unlike with an ordinary electrical capacitor. Contrary to what is stated in the book (p. 254), accurate values of interfacial tension by the 'drop time' method are not obtainable even with the corrections (for back pressure and incomplete dislodgement of the mercury drop). This is due to the fact that the capillary bore is wetted by the electrolyte solution and one observes a hysteresis of the contact angle at the mercury-solution interface inside the capillary.

In eq. II, 50, the concentration terms should be defined properly as those prevailing adjacent to the electrode surface. Eq. II, 77–79 are again misleading with respect to the concentration term : C° should be defined as at the outer edge (solution side) of the diffuse double layer or as the mass-transfer controlled value at the electrode surface. Otherwise, confusion may be caused later, for example between eq. II, 80 and eq. II, 77.

Notwithstanding these remarks, a book such as this is welcome to the electrochemical literature since there is a paucity of information on conducting laboratory experiments in interfacial electrochemistry.

S. Satyanarayana.

Nuclear and Particle Physics. Part A. Background and Symmetries. By Hans Frauenfelder, and Ernest M. Henley. (W. A. Benjamin, Inc., Reading, Massachusetts 01867, U.S.A.), 1975. Pp. xviii + 573. Price : Cloth : \$ 21.50 ; Paper : \$ 13.50.

This is a graduate level text on nuclear and particle physics. These two areas are concerned primarily with short range forces and high energies. Any treatment of these has to perforce emphasize general symmetry considerations to extract generally valid results and the Part A, under review, is devoted to these and general background material.

The treatment of the symmetries covered which include geometric conservation laws, isospin parity, charge conjugation, Time-Reversal, TCP is rather leisurely and instructive. The examples are drawn both from the field of elementary particle physics and low energy physics. Thus the discussion of isospin ranges from evidence from low energy nuclear physics, isospin for nuclei, isobaric analog state, displaced charged multiplets for elementary particles and particle reactions, isospin in field theory. This feature of the book, which is expected from the title of the book, is especially welcome in these days of excessive fragmentation of physics.

Most books on symmetries imply a direct relationship between a symmetry and a conservation law. Thus, it is generally stated that the time displacement invariance of fhe physical laws leads to the energy conservation (*see* page 33 or 56). This is true if the dynamics obeys a lagrangian principle but this is not true in general. For example, the equation of motion of a particle moving in a viscous medium $m\ddot{x} + k\dot{x} = 0$ while invariant under $t \rightarrow t +$ constant does not obviously admit of a conserved quantity like "Energy". In this case, of course, one cannot write a lagrangian which explicitly does not depend on time. This is only a note of caution.

I found, on the whole, the book to be excellently written. It can be used for self-study by an aspiring student who has had a course in quantum mechanics. A large number of exercises add to the value of the book. There are a number of delightful historical and other asides in the book which add to the enjoyment of reading the book. One would eagerly wait for the second part of the book which will deal with electromagnetic and weak interactions of nuclei and particles. VIRENDRA SINGH.

Population and Community Ecology. By E. C. Pielou. (Gordon and Breach, London, W.C. 2), 1975. Pp. 424. Price $ 39.00.

This sequel to Pielou's 'Introduction to Mathematical Ecology' is a most welcome addition to the rapidly growing ranks of modern texts on ecology. It contains a very sound exposition of ecological theory developed along the non-evolutionary point of view. This viewpoint accepts the various ecological parameters such as birth and death schedules or competition coefficients as given, and works out their consequences for the dynamics of populations or communities. The question of why these parameters come to assume particular values is set aside.

The book is aimed at the biologist with little mathematical background, and develops most of the required mathematical tools in the appendices to the main text. This approach has succeeded admirably, and the book should provide a very good introduction for the non-mathematical biologist to the increasingly sophisticated body of ecological theory.

The theoretical exposition is well supplemented with abundant illustrations from the real world and serves to give a good feel for the various facets of ecology. Portions of the book dealing with age structure, spatial patterns and ecological diversity make for particularly enjoyable reading. It is a pity that the very high price of the book would make it quite impossible for any individual in our country to acquire it. MADHAV GADGIL.

Horizons in Biochemistry and Biophysics (Vol. 1), Edited by E. Quagliariello, F. Palmieri and T. P. Singer. (Addison-Wesley, Advanced Book Programme, Reading, Massachusetts, USA), 1974. Pp. xiii + 344. Price $ 13.50.

This volume is the first in the series "Horizons in Biochemistry and Biophysics" and is therefore an experimental one. The professed objective of the editors is to improve communication among scientists and students (Sic !). The first of the nine reviews is on "Superoxide and Evolution" by Fridovich. The subject has been treated at a level that should attract both the novice and the expert. Kenney's chapter on the 'Molecular Nature of Isozymes' is well written. While the volume, as a whole, cannot be accused of catering solely to the esoteric, the chapters on NADH pathways and Flavin coenzymes do tend to get very specialised.

"Ferredoxins and Photosynthesis" is a good exposition, by Arnon and Buchanan, of a subject which has attracted a great deal of attention, recently. This article does not contain a summary, unlike the earlier ones. Coverage of other topics like 'abnormal enzymes', 'membrane fluidity', 'metabolic regulatory functions of oxalacetate, and 'coupling site I' is adequate. The editors are to be congratulated for largely preserving the lucidity of the presentation. They have made their excuses in the preface, but one cannot help noticing their marked preference for the Veterans Administration Hospital, San Francisco. Five of the nine reviews originate from that worthy institution and the editors' claim, that scientists from two continents are represented, is hard to support. It is to be hoped that future volumes will be more truly representative in the choice of contributors, as well as subjects.

P. BALARAM.

Black Holes, Gravitational Waves and Cosmology. By R. Rees, R. Ruffini, and O. A. Wheeler. (Gordon and Breach, Science Publishers, New York), 1974. Pp. xvi + 166. Price £ 12.80.

In the last few years, very important developments have taken place in Astrophysics and Cosmology. These include Pulsars, Black Holes, Gravitational

Waves and Microwave Background Radiation. These are intimately connected with the evolution of the universe. There are two rival theories on Cosmology at the moment. One theory starts with the assumption that the universe in the beginning was a highly condensed hot primordial fire ball which exploded with a big bang. The Galaxies, Stars and Planets subsequently evolved in the expansion process following the big bang. One of the evidences in support of this theory is Microwave Background Radiation. The second model is the steady states theory and suggests that the universe has neither a beginning nor an end. However, one has to visualise creation of matter to explain the uniform matter density.

The concept of black holes which has been worked out in detail recently has led to exciting developments such as the Kerr Holes, the Schwarzschild Black Holes and Kerr-Newman Black Holes. In fact, as have beon shown by the reviewer and his collaborator (C. Sivaram), these concepts may have connection with elementary particles through formation of Quantum black holes. Thus the study of black holes might throw light on the basic building blocks of matter (*i.e.*. elementary particles). Another exciting development is the attempt to detect gravitational radiation and the source of such radiation. While there are various experimental attempts made to detect the gravitational waves, the field is wide open and considerable experimental and theoretical researches will be required to settle this situation in one way or the other.

The book under review contains all these topics in a very readable style. It is full of information about the recent concepts and experimental results. It has also a chapter on the Large Number Hypothesis and the concluding chapter on "beyond the end of time", deals with the problem of gravitational collapse, which to the authors appear the greatest crisis in physics. The recent work of the reviewer and his collaborator in which the supergravity has been advocated may offer a solution to this and it appears that the microcosmos and the cosmos may be connected. The book also contains a selection of certain reprints of papers published on black holes and gravitational waves. The reviewer finds the book extremely valuable for research workers in this field. It will be useful both to the theorists and experimentalists and the book is strongly recommended.

K. P. Sinha.

Structural Stability and Morphogenesis. By Rene Thom. (W. A. Benjamin, Inc.), 1975. Pp. xxv + 573. Price $ 21.50.

The theory of catastrophes as developed by Prof. Rene Thom has created a great deal of intellectual excitement among mathematicians, physicists and biologists. The reviewer delayed this review for the simple reason that he was awaiting his participation in a conference on Evolutionary Models in Nature in Europe where Prof. Thom himself was to come and put forward his ideas about the theory of catastrophes. He was indeed fortunate to have a first account of this work in December, 1975.

Prof. Thom's work deals with those systems which can be described by a set of finite variables which are in turn controlled by a set of finite variables. He studied the stability of the system when the control variables are changed from the property of the energy function involving both types of variables. Prof. Thom has invoked some elegant ideas of Topology. According to Thom's theorem, with the exception of certain cases, it is possible to affect a smooth reversible change of co-ordinates in such a manner that the system shows one of the 7 types of behaviours. And these 7 types of behaviour are of great importance in various applications. These are known as 7 types of catastrophes depending on the corresponding energy function. These catastrophes are named as Fold, Cusp, Swallowtail, Butterfly, Hyperbolic umbilic, Elliptic umbilic and Parabolic umbilic. For some critical values of controlled variables, this system can go from one stable structure to another stable configuration by what is called as a catastrophic jump. This theory of catastrophe is finding application in the collapse of a bridge, optical catastrophes, catastrophic flow of fluids and biological systems. There may be catastrophic changes in evolving biological systems. The theory has also applications to social problems and in dramatic economic changes.

The book under review is essentially mathematical in nature although Prof. Thom, has tried to explain it with many pictorial diagrams and representations. A person trying to understand this subject has to learn the necessary mathematical techniques such as Topology. Prof. Thom has addressed himself mainly to classical systems. It is not clear to the reviewer whether his theory can be applied to quantum systems and, in particular, to systems like superfluids, superconductors and magnetic systems undergoing phase transitions.

There is no doubt that this theory of catastrophe as given by Prof. Thom, will find enormous applications in various branches of science, namely, physics, biology and sociological models. The reviewer strongly recommends this to the scientists trying to do research in these interdisciplinary areas.

K. P. Sinha.

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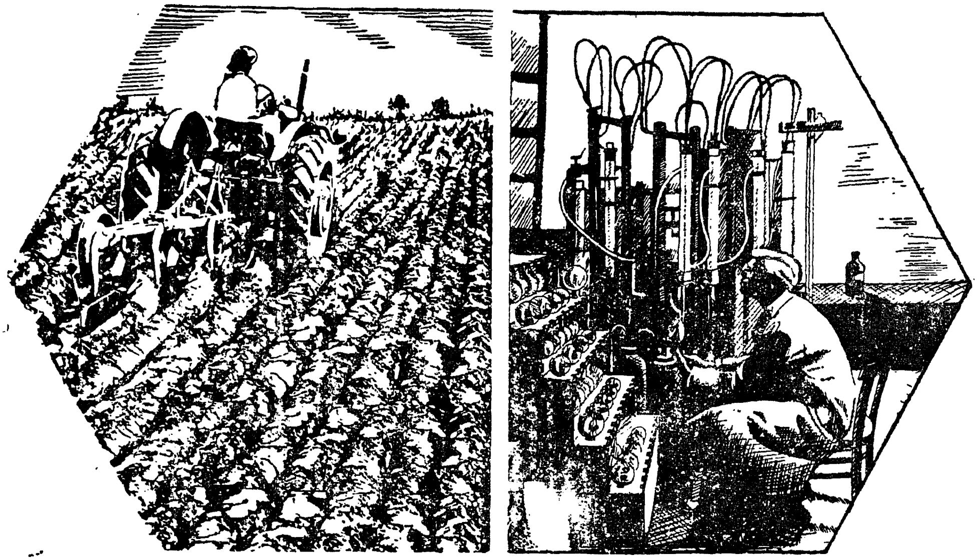

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MÖSSBAUER AND INFRARED SPECTRAL STUDIES ON THE COMPLEXES OF SOME ALKALOIDS WITH FERRO- AND FERRICYANIC ACIDS

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ABSTRACT

The reactions of alkaloids like quininine, cinchonine, quinaldine and procaine with ferro- and ferricyanic acid have been reported. Procaine forms a 2 : 1 complex with ferricyanic acid and 1 : 1 complex with ferrocyanic acid while all other alkaloids give 1 : 1 complexes. Infrared studies show that the alkaloids undergoes protonation of the amine group during complex formation. The C ≡ N stretching frequency appears as a doublet which may be explained on the basis of unsymmetrical hydrogen bonding. Mössbauer studies have shown that iron in hydroferro- and ferrisyanide complexes is present as Fe^{2+} and Fe^{3+} respectively. The isomer shift and quadrupole splitting shows the slight distortion in the symmetry hexacyanoferrate (II) and (III).

INTRODUCTION

REACTIONS of complex iron cyanides with organic bases and related compounds have been reported[1,2]. However, very limited studies have been carried out with an important class of compounds, the alkaloids. Only recently Burkat[3] reported the reaction of antipyrine in acidic medium with potassium hexacyanoferrate (II) while Kreshkov *et al.*[4,5] reported the reaction of dimethylamino antipyrine with potassium hexacyanoferrate (III). Since, nothing much has been done in this direction, studies have been carried out on the reaction of some alkaloids with ferro- and ferricyanic acids.

EXPERIMENTAL

Reagents :

Ferro- and ferricyanic acids were prepared from potassium hexacyanoferrate (II) and (III) (AnalaR grade, BDH) by the method of Brauer[6,7]. The alkaloids, quinine sulphate, cinchonine, quinaldine and procaine hydrochloride were of reagent grade and purified by crystallisation from ethanol or double distillation. All other reagents were AnalaR grade (BDH).

Conductometric titrations were performed on Toshniwal Conductivity Bridge CLO1/OIA (India) using a lip type conductivity cell. Infrared spectra of the complexes were recorded on a Beckmann IR-20 infrared spectrophotometer in KBr discs. Mössbauer studies were carried out on Mössbauer spectrometer MBS 35 (ECIL, Hyderabad) using a Co^{57} source in Rh matrix having an initial activity of 2 mCi. The spectrometer was calibrated against sodium introprusside.

Preparation of the Complexes :

The complexes were prepared by mixing in stoichiometric ratio, a 0·01 M solution of ferro- or ferricyanic acid with a 0·01 M solution of the alkaloid in alcohol. The precipitate formed was filtered off at once and washed with alcohol until all the excess unreacted reactants are removed. The precipitate was then dried in vacuum over anhydrous calcium chloride.

Chemical Analysis :

Iron was estimated spectrophotometrically using 1, 10 phenanthroline after decomposing the complexes with aqua regia. Water of crystallisation was determined by desiccating the complexes at about 100° C. Carbon, hydrogen and nitrogen were estimated at I.I.P., Dehradun (India).

RESULTS AND DISCUSSION

The composition of the complexes was determined by conductometric titrations, which shows that procaine forms a 2 : 1 complex with ferricyanic acid and 1 : 1 complex with ferrocyanic acid. On the other hand, quinine, cinchonine and quinaldine form 1 : 1 complexes both with ferro- and ferricyanic acids. The composition of these complexes, as determined by chemical analysis are given in Table I.

Infrared Studies :

The infrared spectra (Table II) of those complexes show O–H symmetric and asymmetric stretching bands and H–O–H bands in the range 3360–3400 cm^{-1} and 1600–1650 cm^{-1} respectively, indicating the presence of water of crystallisation. The Fe–C stretching band in the hydroferro- and ferricyanide complexes appears in the range 520–600 cm^{-1}.

The C ≡ N stretching frequency appears in the range 2000–2070 cm^{-1} for the hydroferrocyanide complexes and 2020–2140 cm^{-1} for the hydroferricyanide complexes. The appearance of a doublet for the C ≡ N frequency is explained on the basis of protonation of alkaloid and subsequent hydrogen bonding[9, 10] as proposed for ferro- and ferricyanic acids.

TABLE I

Elemental analysis of the complexes

Complex	H_2O%		Fe%		C%		N%		H%	
	Cal.	Found	Cal.	Found	Cal.	Found	Cal.	Found	Cal.	Found
1. $H_4[Fe(CN)_5]$ $[C_{20}H_{24}N_2O_2]$ 0·5 H_2O	1·63	2·00	10·20	10·16	56·83	57·95	20·40	21·01	4·91	4·75
2. $H_3[Fe(CN)_6]$ $[C_{29}H_{24}N_2O_2]$ 1·0 H_2O	3·23	3·15	10·05	10·27	56·01	57·32	20·10	21·35	5·20	5·45
3. $H_4[Fe(CN)_6]$ $[C_{19}H_{22}N_2O]$ 1·0 H_2O	3·40	3·61	10·60	10·89	56·81	55·76	21·21	20·97	5·30	5·54
4. $H_3[Fe(CN)_6]$ $[C_{19}H_{22}N_2O]$ 0·5 H_2O	1·73	2·01	10·81	10·99	57·91	57·02	21·62	20·98	5·01	5·39
5. $H_{66}[Fe(CN)]$ $[CH_3CH_{91}N]$ 0·5 H_2O	2·44	2·54	15·21	15·01	52·17	53·07	26·63	27·39	3·80	3·62
6. $H_3[Fe(CN)]$ $[CH_3C_1H_1N]$ 1·0 H_2O	4·78	4·98	14·89	14·57	51·06	51·89	26·06	26·79	3·72	3·94
7. $H_4[Fe(CN)_6]$ $[C_{13}H_{26}N_2O_2]$ 0·5 H_2O	1·95	2·27	12·14	11·96	49·45	50·75	24·29	25·15	5·42	5·71
8. $H_3[Fe(CN)]$ $[C_{93}H_{22}N_2O_2]$ 0·5 H_2O	1·29	1·60	8·04	8·20	55·17	56·17	20·11	20·99	6·32	6·21

TABLE II

Infrared absorption bands of the complexes (Cm^{-1})

Complex	$\nu(H_2O)$	ν(NH), $\nu(NH_3^+$	$\nu(C\equiv N)$	NH^+, NH^+_3	$\nu(Fe-C)$
Quinine hydroferrocyanide	3400 (b, st) 1620	2600 (b) $\nu(NH^+)$	2000 (s, st) 2020 (s, st) 2040 (sh)	1390 (s) (NH^+)	570 (s, st)
Quinine hydroferricyanide	3400 (b,st) 1610	2600 (b) $\nu(NH^+)$	2100 (s, st) 2140 (s, st)	1390 (s) (NH^+)	520 (st)
Cinchonine hydroferrocyanide	3360 (b) 1620 (m)	2650 (w) $\nu(NH^+)$	2030 (s, st) 2060 (s, st)	1730 (s) (HN^+)	540 (b)
Cinchonine hydroferricyanide	3380 (m) 1625 (w)	2700 (w) $\nu(NH^+)$	2080 (s, st) 2120 (sh)	1400 (w) (NH^+)	600 (w))
Quinaldine hydroferroeyanide	3400 (b) 1650 (m)	—	2010 (sh) 2070 (s, st)	1390 (s) (NH^+)	550 (st)
Quinaldine hydroferricyanide	3400 (b) 1630 (m)	2600 (w) $\nu(NH^+)$	2080 (s) 2110 (s, st)	1395 (s) (NH^+)	590 (m)
Procaine hydroferrocyanide	3400 (b) 1610 (b)	2840 (w) $\nu(NH_3^+)$ sym	2050 (s, st)	1470 (w) (NH_3^+) sym	560 (s, st)
Procaine hydroferricyanide	3400 (b) 1605 (s)	2960 (w) $\nu(NH_3^+)$ asym	2020 (sh) 2100 (s, st)	1550 (w) (NH_3^+) asym	620 (w)

s = sharp, st = strong, sh = shoulder, b = broad, m = medium, w = weak.

The N–H symmetric and asymmetric stretching bands and N–H deformation bands of the alkaloids shifted to a lower frequency[11-13], on complex formation which indicate protonation of the alkaloids.

In the light of the above considerations it may be concluded that the interaction of the alkaloid with ferro- and ferricyanic acids is taking place through unsymmetrical hydrogen bonding between

TABLE III

Mössbauer parameters of hydroferro- and ferricyanide complexes

Sl. No.	Alkaloid	Hydroferrocyanides		Hydroferricyanides	
		Isomer shift δ (mm/sec) ±0·02	Quadrupole splitting $\triangle E_q$ (mm/sec) ± 0·02	Isomer shift δ (mm*t*sec ±0·02)	Quadrupole splitting $\triangle E_q$ (mm*t*sec) ± 0·02
1.	Quinine	−0·206	No	−0·268	0·845
2.	Chinchonine	−0·232	No	−0·263	0·834
3.	Quinaldine	−0·223	No	−0·227	1·133
4.	Procaine	−0·227	No	−0·278	0·824
	Hexacyano-ferrate (II)	−0·138	..	..	..
	Hexacyano-ferrate (III)			−0·186	0·132

the nitrogen of the cyanide group and proton of the amine group of the alkaloid.

Mössbauer Studies :

The Mössbauer spectra of the hydroferrocyanide complexes (Table III) show a small isomer shift and on quadrupole splitting. On the other hand small quadrupole splitting and small isomer shifts are obtained in the case of hydroferricyanide complexes.

The values of isomer shift and quadrupole splitting show that iron in hydroferro- and ferricyanide complexes is present as Fe^{2+} and Fe^{3+} respectively[14]. The low values of isomer shift also indicate that they are low spin complexes.

The values of isomer shift and quadrupole splitting in these complexes show a change from those of hexacyanoferrate (II) and (III). This change may be attributed to slight distortions in the symmetry of hexacyanoferrate (II) and (III) on interaction with the alkaloid due to unsymmetrical hydrogen bonding which supports the conclusions drawn on the basis of splitting of the $C \equiv N$ stretching frequency in the infrared spectra.

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1. Gadreau, H., *J. Pharm. Chim.*, 1929, 6, 145.
2. Cumming, W. M. and Stewart, J. A., *J. Soc. Chem. Ind.*, 1956, 51, 273-6T.
3. Burkat, S. E., *Aptechone Delo,* 1956, 5 (3), 26.
4. Kreshkov, A. P., Vilborg, S. S., Filippova, K. I. and Drogdov, V. I., *J. Anal. Chem., U.S.S.R.*, 1956, 11, 215.
5. —, —, — and —, *Zhur. Anal. Khim.*, 1956, 11, 212.
6. Brauer, G., *Hand Book of Preparative Inorganic Chemistry,* Academic Press, New York, 1965, 2, 1509.
7. —, *Hand Book of Preparative Inorganic Chemistry,* Academic Press, New York, London, 1961, p. 787.
8. Vogel, A. I., *A Text Book of Quantitative Inorganic Analysis,* 3rd Ed, Longmans Press, 1961, p. 787.
9. Garg, A. N. and Goel, P. S., *J. Inorg. Nucl. Chem.*, 1969, 31, 697.
10. — and —, *Ibid.*, 1970, 32, 1547.
11. Chenon, and Sandorfy, *Canad. J. Chem.*, 1958, 36, 1181.
12. Mitchell, P. C. H., *J. Inorg. Nucl. Chem.*, 1961, 21, 382.
13. Greenwood, N. N. and Wade, K., *J. Chem. Soc.*, 1960, p. 1130.
14. Goldanskii, V. I. and Herber, R. H., *Chemical Application of Mössbauer Spectroscopy,* Academic Press, New York, 1968, p. 163.

ACS ELECTION OF CENTENNIAL FOREIGN FELLOWS

Dr. C. N. R. Rao, Professor of Chemistry, Indian Institute of Technology, Kanpur, has been elected as a Fellow from India.

PHYSICO-CHEMICAL STUDIES OF SCHIFF'S BASE METAL COMPLEXES

B. L. WAZIR AND K. P. DUBEY

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ABSTRACT

Stability constants of iron, nickel, cobalt, zinc, copper and manganese complexes with Schiff's bases derived from salicylaldehyde and the amines *p*-aminophenol and *m*-aminophenol, have been determined in 50% ethanol water mixtures at constant ionic strength. The order of stability constast found is

$Cu^{2+} > Fe^{2+} > Ni^{2+} > Co^{2+} > Zn^{2+} > Mn^{2+}$.

Values of $-\triangle F$, δH and Er (Mn – Zn) have been caculated.

SCHIFF'S bases formed by the condensation of an amine with an aldehyde or a ketone have been used for preparing metal complexes. A large variety of such metal complexes has been prepared and their structure determined. Recently some interest has been shown in the metal complex equilibria studies using Schiff's bases as ligands, but in general such studies have been limited to a few metal Schiff's bases and pH titrations are performed to evaluate stabilities of a variety of metal ions with Schiff's bases having oxygen as potential donor. Stability constants of their metal chelates have been determined in 50% alcoholic solutions of ionic strength 0·05 M (KNO_3) at 25° C using Bjerrum calvin pH titration technique[1-2].

EXPERIMENTAL

Preparation of Schiff's Bases:

Schiff's bases of *m*-aminophenol and *p*-aminophenol with salicylaldehyde were prepared by refluxing equimolar quantities of the amine and the salicylaldehyde and were purified by recrystallization from ethanol[3]. Ligand solutions (0·01 M) were prepared in absolute alcohol. Solutions of metal ions (0·005 M) were prepared from B.D.H. analar grade reagents and were standarized by conventional methods[4]. Potassium nitrate and nitric acid were used to maintain ionic strength.

Titration Method:

The following solutions were prepared:

(A) 5 ml KNO_3 (0·5 M) + 10 ml HNO_3 (0·01 M) I
(B) A + 5 ml ligand solution (0·01 M) II
(C) B + 2 ml metal solution (0·005 M) III

The total volume in all the cases was 50 ml, each containing 50% ethanol. The solutions were kept in a thermostat and then titrated separately against standard NaOH solution.

Calculations:

(1) *Protonation constants*: From the titration curves of the solutions (I) and (II) $\bar{n}$H at different pH values were calculated using the method of Irving and Rossotti[5]. Protonation constants were evaluated using Bjerrum's half $\bar{n}$ method and Speakman's method[6]. The values of the protonation constants obtained are given in Table I.

TABLE I

Schiff's base of Salicylaldehyde with	Yield %	Protonation constants of Ligand at 0·05 M ionic strength		
		Log K_1	Log K_2	Log K_3
1. *m*-aminophenol	72	9·90	8·05	4·65
2. *p*-aminophenol	78	10·10	8·16	5·00

Metal Ligand System:

The values of $\bar{n}$ and pL were calculated from the titration curves of the solutions (II) and (III). From the formation curves the values of stepwise stability constants were evaluated using the method of interpolation of $\bar{n} = 0·5$. The δH values were calculated according to the method described by George and McClure[7]. The values of step stability constants of metal chelates and δH are given in Tables II and III.

RESULTS AND DISCUSSION

From the results, it is seen that the order of stability of first row of transition metal complexes (d^5–d^{10}) with Schiff's bases is as follows:

$Cu^{2+} > Ni^{2+} > Co^{2+} > Zn^{2+} > Mn^{2+}$.

Except for the Fe (II) complexes, this sequence is in agreement with the Mellor and Maley[8-9] order. The abnormally high stabilities of Fe (II) complexes can be attributed to the resonance stabilization energy of Fe (II) complexes upon co-ordination with a ligand having aromatic ring systems[10]. The values of δH observed for Co (II), Ni (II) and Cu (II) complexes are also of the same order as for other ligands co-ordinating through

TABLE II

Ligand cations	Schiff's base of salicylaldehyde with *m*-aminophenol			$-\triangle$ F KCal/mole	Schiff's base of salicylaldehyde with *p*-aminophenol			$-\triangle$ F KCal/mole
	Log K_1	Log K_2	Log β_2		Log K_1	Log K_2	Log β_2	
Mn^{2+}	4·70	4·4	9·10	12·41	4·15	—	4·15	5·65
Fe^{2+}	7·15	5·7	12·85	17·53	7·40	—	7·40	10·09
CO^{2+}	6·15	4·8	10·95	14·94	4·35	—	4·35	5·93
Ni^{2+}	6·40	4·9	11·30	15·41	4·45	—	4·45	6·07
Cu^{2+}	10·30	6·8	17·10	23·32	9·20	—	9·20	12·55
Zn^{2+}	5·3	5·1	10·40	14·19	5·50	—	5·50	7·50

TABLE III

Calculations of Er (Mn–Zn) and of δH values for complexes of Schiff's base from salicylaldehyde and m-*aminophenol*

Cations	Mn^{2+}	Fe^{2+}	Co^{2+}	Ni^{2+}	Cu^{2+}	Zn^{2+}
Long K_1	4·7	7·15	6·15	6·4	10·3	5·3
$\triangle$ GK Cal/mole	6·41	9·752	8·389	8·73	14·05	7·23
$\triangle H_c$ (*i.e.*, $\triangle$ G relative to Mn^{2+} value)		3	2	2	8	1
$\triangle H_H$ (relative to Mn^{2+} value)		26	43	62	63	47
$\triangle H_L$ (relative to Mn^{2+} value)		29	45	64	71	48
$[(n-5)/5]\ E_r$		10	20	30	40	—
δ H		19	25	34	31	—

$\triangle G$—Free energy change = 2·303 RT log K (where RT and K have the usual significance; T = 300°K).

$\triangle H_c$ — Change in heat content for the formation of the complex in solution.

$\triangle H_H$ — Heat of hydration of metal ion.

$\triangle H_L$ — Heat of complexation referred to metal ion in gaseous and ligand in solution state.

n — Number of electrons in 3rd orbital.

E_r — Lattice energy difference for Zn^{2+} and Mn^{2+} complexes.

δH — Thermodynamic stabilization energy.

one nitrogen atom. This confirms that the metal ions are bonded through one nitrogen and one oxygen atom to the ligand molecule.

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1. Bjerrum, J., *Metal Ammine Formation in Aqueous Solution*, P. Haase and Son, Copenhagen, 1941.
2. Calvin, M. and Wilson, K. W.., *J. Amer. Chem. Soc.*, 1945, 67, 2003.
3. Katiyar, S. S., Raut, K. B. and Arvindakshan, P. A., *Curr. Sci.*, 1969, 38, 242.
4. Vogel, A. I., *Quantitative Inorganic Analysis*, Longmans, London, 1962.
5. Irving, H. M. and Rossotti, H. S., *J. Chem. Soc.*, 1954, p. 2904.
6. Speakman, J. C., *J. Chem. Soc.*, 1940, p. 855.
7. George, P. and McClure, D. S., *Inner Orbital Splitting, Prog. Inorg. Chem.*, Interscience, New York, 1959, 1, 428.
8. Mellor and Maley, *Nature*, 1947, **159**, 370.
9. — and —, *Ibid.*, 1948, 161, 436.
10. Irving, H. and Williams, R. J. P., *J. Chem. Soc.*, 1953, p. 3206.

COLD-TOLERANCE IN THE TENT CATERPILLAR *MALACOSOMA AMERICANUM* AND THE EFFECTS OF HORMONES

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ABSTRACT

The chill-coma temperature and the supercooling point of the third instar larvae of *Malacosoma americanum* were 7·0° C ± 0·4° C and − 14·5° C ± 1·5° C respectively. Treatment of these larvae with 1 or 4 μl of ecdysterone or farnesyl methyl ether had no significant effect on these cold-tolerance values, though there is some indication that the growth hormone may reduce cold-tolerance by raising supercooling point.

The chilled larvae could regain locomotory activity within 8 minutes of being transferred to 30° C from 3 weeks of chilling at 5° C. The hormones had no apparent effect in the acceleration of recovery.

INTRODUCTION

HORMONAL regulation of various life processes in insects has been demonstrated in many species. Diapause or arrest of growth and development is also attributed to the lack of brain hormone, ecdysone or juvenile hormone[1]. However, the control of other forms of dormancy such as quiescence and oligopause is still not clear[2].

Topical application of farnesyl methyl ether (FME) was found to increase cold-tolerance in the larvae of the wax moth *Galleria mellonella* L.[3]. The time required for recovery from quiescence and the overall mortality, after 1 to 3 weeks of chilling at 4° C, were significantly reduced. The results were explained on the basis of the effects of the chemical on the general metabolism in lepidopterous larvae.

In the present study, the possible effects of ecdysone and FME on the chill-coma temperature, as defined by Mutchmor and Richards[4], supercooling point and temperature acclimation were investigated on the larvae of the eastern tent caterpillar *Malacosoma americanum* (Fab.) (Lasiocampide, Lepidoptera).

The biology of *M. americanum* is well suited for the present study. The species is found throughout southern belt of Eastern Canada, and almost all over United States, east of Rocky Mountains[5]. The overwintered fully mature embryos hatch in early May and complete larval development within 5–6 weeks. The adult females lay eggs in early July and the embryonic development takes about 3–4 weeks.

MATERIALS AND METHODS

The tent caterpillars for this study were collected from wild cherry trees around Kingston (Ontario) and reared in the laboratory at 30° C as described by Mansingh[5].

Third instar larvae were either injected with 1 or 4 μl of ecdysterone (supplied by Rohoto Pharmaceuticals, Osaka, Japan) as 10% ethanol solution or topically treated with 1 or 4 μl of FME (supplied by Flunkaag, Buchs, Switzerland). The control larvae received ethanol injection or acetone application. One day after the hormonal treatment, the larvae were transferred to 5° C for 3 weeks, or maintained at 30° C. At the end of the chilling period, the chilled larvae were brought back to 30° C and the time for their recovery from cold-torpor was recorded according to Mansingh[3]. The growth rate during the chilling was determined by weighing the larvae before and after cold-exposure.

The chill-coma temperature was determined by exposing the larvae from 30° C to a series of low temperatures for 1 hour. The temperature at which they became inactive was taken as the chill-coma temperature. Supercooling points were measured with 32-gauge copper-constantan thermocouple while the rate of cooling was 2° C/minute[5].

RESULTS AND DISCUSSION

The amounts of ecdysone or FME used in the present investigation had no apparent effect on the larval development at 30° C or 5° C. Cold exposure had induced torpor within half an hour, and no feeding activity was noticed during the 3 weeks of chilling period. Both the treated and controlled larvae lost about 8·25% of their original weight of 0·26 g, during the cold exposure.

Hormonal treatment had no effect on the chill-coma temperature of the larvae which remained unchanged at around 7·0° (Table I). Similarly, the supercooling ability of the larvae was also not significantly altered by the hormones, though higher dose of ecdysone (4 μl/larva) appear to reduce cold-tolerance by slightly increasing the supercooling point. This is not surprising because chilling and ecdysone treatment have antagonizing effects on the metabolism. Low temperature treatment reduces metabolism and biosynthetic activities[2,6]; ecdysone, on the other hand, stimulates RNA and protein syntheses[7]. This aspect merits further investigation.

TABLE I

Effect of hormones on the cold-tolerance of M. americanum

Treatment	Chill-coma temperature (°C)	Supercooling points (°C)
Control	7·0±0·4	−14·5±1·5
Ecdysone — 1 μl	6·8±0·3	−15·2±1·1
Ecdysone — 4 μl	6·5±0·4	−15·5±0·5
FME 1 μl	7·5±0·2	−13·8±1·0
FME 4 μl	7·8±0·5	−12·8±0·8

20–30 larvae/experiment.

Table II shows that the locomotory activity in the chilled larvae was regained within 10 minutes (8 ± 1·25 minutes) of their return to 30° C. FME or ecdysone had no apparent effect on the recovery time.

TABLE II

Effect of hormones on the recovery of locomotory activity in M. americanum *larvae exposed to* 25° C *after* 3 *weeks of chilling at* 5° C.

Treatment	% larvae recovered 4	6	8	10 (min)
Control	25	45	84	100
Ecdysone — 1 μl	20	40	90	100
Eceysone — 4 μl	35	50	100	100
FME 1 μl	28	45	90	100
FME 4 μl	34	52	88	100

The anomaly in the role of FME in the induction of cold-tolerance in *M, americanum* and *G. mellonella*[3] may be due to the inherent differences in the level of cold-tolerance in the two species. This is evidenced by the ability of tent caterpillar larvae to recover from cold-torpor within 10 minutes, even when the chilling induced irreversible and fatal injury in over 60% of the larvae[5]. The wax moth larvae, on the other hand, required about 15 to 40 minutes to recover, depending upon the duration of chilling.

Intraspecific differences in the two species is further indicated by significant differences, in their chill-coma temperatures. The low chill-coma temperature of the tent caterpillar is almost equal to that of *Musca domestica* or *Sarcophaga bullata;* for the larvae of *G. mellonella,* it is 8·8° C[4]. Mutchmor[6] has discussed the positive correlation between the chill-coma temperature, muscle apyrase activity, ATP-ase activity and cold-tolerance in a number of species ; the lower the chill-coma temperature, the more cold-tolerant is the species. Thus the inherently high cold-tolerance in the tent caterpillar can apparently not be further increased by the low doses of FME.

Increased cold-tolerance and the ability to recover quick from cold-torpor have adaptive significance for the tent caterpillar. It is not unusual for the insect to experience temperatures which fluctuate between 0 and 25° C ; the wax moth larvae rarely face such adversity. Consequently, *M. americanum* larvae have evolved the ability to adapt quickly to the overnight low temperatures, prolonged spells of cold or high day temperatures.

ACKNOWLEDGEMENTS

Prof. B. N. Smallman, Department of Biology, Queen's University, Kingston. Ontario, Canada, had kindly provided the laboratory facilities, and the U.W.I., the travel grant.

1. Wigglesworth, V. B., *Advances in Insect Physiology,* Ed. Beament, J. W. L. *et al.*, Academic Press, New York, 1964, 2, 248.
2. Mansingh, A., *Can. Ent.,* 1971, 103, 983.
3. —, *Can. J. Zool.,* 1973, 51, 61.
4. Mutchmor, J. A. and Richards, A. G., *J. Insect Physiol.,* 1961, 7, 141.
5. Mansingh, A., *Can. J. Zool.,* 1974, 52, 629.
6. Mutchmore, J. A., *Amer. Assoc. For Advance of Sci. Washington, D.C.,* 1967, p. 165.
7. Sahota, T. S. and Mansingh, A., *J. Insect Physiol.,* 1970, 16, 1649.

ANTHOLYSIS OF *SESAMUM INDICUM* L. (TIL)

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ABSTRACT

Antholysis leading to virescene, phyllody and prolification of *Sesamum indicum* has been studied and described here. Infected flowers developed chlorophylls and mode carotenoids; but the leaves showed lower (mg/g) chlorophylls as well as carotenoids, when compared to healthy plants.

INTRODUCTION

ANTHOLYSIS (Gr. anthos-flower; lysis = loosing, dissolution) is termed as abnormality progressing from a normal flower to a completely vegetative leafy branch. It entails virescence, phyllody, and prolification. Antholysis starts with *virescence* or *greening* of normally white or coloured parts by the development of chloroplast[1]. Green flowering diseases[2], phyllody[3], witches broom[4], phenomena leading to complete sterility have been indicated on *Sesamum indicum*. A severe antholysis of *Sesamum indicum* appeared in the Til crop in early August 1975, at Jodhpur.

OBSERVATIONS

The Til crop sown in the first week of June 1975, grew well, passing through an early dry phase and later on more moist conditions due to persistent rainfall which is unusual in Indian arid zone. Suddenly in the first week of August some distinct and peculiar virescence in tops of some plants 80–120 cm tall often drooping, made its appearance. On closer examination it was discovered that antholysis has led to the virescence, *i.e.*, turning of pink and purplish-pink coloured flowers into completely green, thickened and puckered ones (Fig. 1). This infection appears to have been transmitted on the tops of the plants because these infected inflorescences were found to have normal healthy fruits at the lower nodes (Fig. 2), but ultimately causing phyllody, *i.e.*, development of floral parts as almost normal foliage. In extreme cases, the flower turned into vegetative shoot (Fig. 3). The infection appeared to have definitely travelled basipetally because: (*a*) Lower portions have normal flowers and fruits which gradually got acropetally infected (Fig. 2); (*b*) The healthy fruits when received the infection burst out and the immature white ovules started to grow into small projected papillae (Fig. 4); and (*c*) When the infected tops were removed the infection made its appearance in the lower axillary branches which developed after a few days.

The effect of antholysis on chlorophyll *a*, chlorophyll *b* and carotenoids[5] is given in Table I. The optical densities of acetone soluble pigments at different wavelengths is shown in Fig. 8. Some of the remarkable features which have appeared after this infection are as follows:

1. The general appearance of the infected plant was characteristic due to green colour and distinctly drooping inflorescences which were visible from a distance (Figs. 5 and 8).

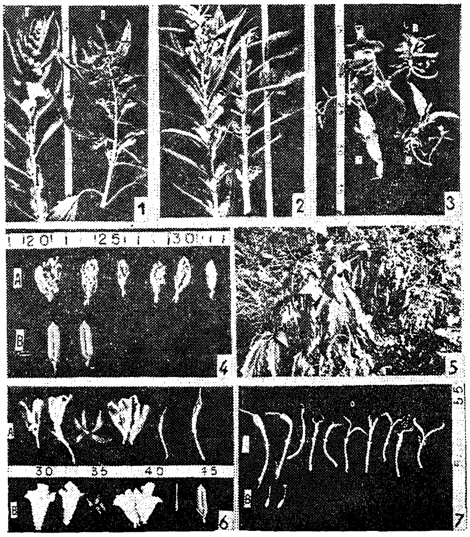
2. Starting from lower levels there were healthy fruits above them some later infected ones which got burst, followed still higher by flowers, pale-green in colour with lesser or no transformation where the petals had shrivelled. On the top came the region where the sudden and total transformations of the different parts of the flowers had taken place. This is what has been termed as antholysis (Fig. 2).

3. The pedicel has elongated successively from lower levels to the higher; sepal elongated with distinct innervation. The petals became green, leathery and puckered (Fig. 6). The number of stamens became reduced to four with distinct sterility, some of the stamens did not bear anthers and often the anther portion became leaf like. The ovary became extremely modified in different ways assuming a leafy appearance (Fig. 7). The lower flowers had enlarged flattened ovary with reduced style which became distinctly vascularised and remained persistent. It also had a tendency to become leaf like.

4. In extreme cases at the top, although there was a tendency towards overall reduction in size of different parts of the flower, but an extreme tendency of conversion of all floral parts into leaf like structures, *i.e.*, phyllody was noticed. There were cases when there was no difference in appearance of sepal, petal, stamen and carpel except that they varied slightly in size.

5. The absolute nature of bilipped corolla is transformed into funnel shaped one and ultimately to rotate type. The petals got separated very much giving a polypetalous appearance. The latter types of flowers show extreme cases of sterility both of androecium and gynoecium. A few pollen grains which were found in infected flowers were empty and without any cytoplasmic contents.

6. An absolute case of vegetative nature of the flower was seen when the ovary got transferred into an axis which ultimately bore leaves and flowers. This was a case of vegetative shoot arising from a flower (Fig. 3).



FIGS. 1–7. *S. indicum*. Fig. 1. infected (A) and healthy (B) shoots, showing difference in flowers. Fig. 2. infected shoot showing grades of infection from lower to upper nodes. Fig. 3. Extreme cases of infection, where transformation of flower into vegetative shoot has taken place. Fig. 4. Healthy fruits (B) after receiving the infection burst out successively (A). Fig. 5. The heavily infected shoot of *S. indicum* plant with characteristic drooping inflorescence. Fig. 6. The infected (A) flowers (intact) calyx, corolla, gynoecium and fruit (all dissected out), compared with their healthy (B) counter-parts. Fig. 7. Different stages of infected (A) ovaries assuming leafy appearance.

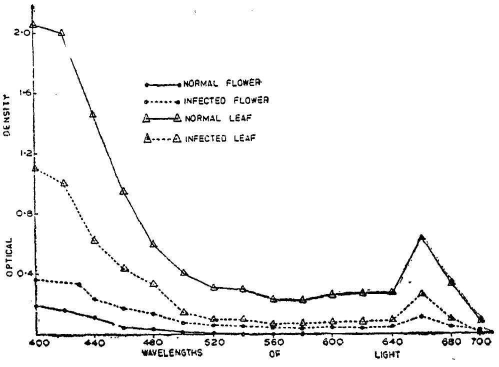


FIG. 8. Optical densities of acetone soluble pigments in healthy and infected leaves and flowers at different wavelengths of light.

7. The petals which are normally caducous in healthy plants transferred into leathery ones and did not fall, remaining persistent enclosing a deformed fruit as they were photosynthetic.

8. The cut parts of a healthy plant which produce mucilage when kept in water, did not show this feature in infected ones.

9. Although the plants showed heavy infection in the floral parts which tends to travel in the basipetal order, surprisingly, did not effect the leaves morphologically although it did effect the pigment system (Table I). However, the leaves at top were slightly reduced.

TABLE I

Chlorophylls a, b, *total chlorophylls and carotenoids mg/g fresh weight in infected and healthy flowers and leaves of* S. indicum

Plant material	Chlorophyll *a*	Chlorophyll *b*	Total chlorophylls	Carotenoids
Healthy flower	0	0	0	0·0575
Infected flower	0·4279	0·1373	0·5652	0·2376
Healthy Leaf	2·5665	0·4770	3·0435	2·8767
Infected Leaf	1·0550	0·1624	1·2174	0·5830

It is evident that the infection appeared to have been transmitted by some insect at a very late stage of plant growth. Contamination was not caused by contact of infected plants with the healthy ones. Artificially, when the extract of an infected plant was rubbed over the healthy plants, they did not receive the infection in this manner. The seeds of *S. indicum* when kept for germination in petri-dishes under controlled conditions with different dilutions of the extract of infected flowers, germinated normally, with only slight curvature in cotyledonary leaves. The seedlings so obtained when grown in pots under natural conditions did not show any sign of infection at all. Thus, it appears that the infection is caused in a very precise and specific way and only means of control at this stage appears to be uprooting of plants.

We wish to thank Prof. Dr. H. C. Arya, for facilities and to the authorities USDA, for providing funds for carrying out investigations on Til under a PL-480 Project to one of us (DNS) during which these observations were made.

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1. Bos, L., *Symptoms of Virus Diseases in Plants*, Oxford and IBH Publ. Co., New Delhi, 1970.
2. Pal, B. P. and Nath, P., *Indian J. Agric. Sci.*, 1933, 5, 517.
3. Robertson, H. F., *Rep. Mycol. Burma* (abstr. in *Rev. Appl. Mycol.*, 1929, **8**, 355.
4. Roy, S. C., *Agriculture Live-stk. India*, 1931, 1, 282.
5. Arnon, D. I., *Plant Physiol.*, 1949, **24**, 1.

EMBRYOLOGICAL STUDIES IN *LAGENARIA SICERARIA* STANDL.

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ABSTRACT

The embryological studies of *Lagenaria siceraria* revealed that the pre-fertilization and post-fertilization growth stages followed the characteristic pattern of development of the Cucurbitaceae family. The development of the female gametophyte follows polygonum type. The development of endosperm was nuclear type and the seed coat had its origin from the integument.

INTRODUCTION

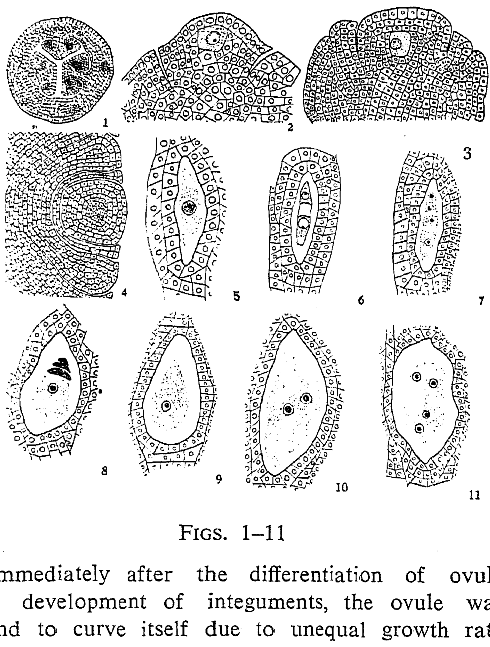
LAGENARIA SICERARIA, commonly known as the bottle gourd, is a monoecious species. In India its green fruits are used as vegetable. Chakravarty (1946) described the morphology of the genus *Lagenaria* and its species. Kirkwood (1905) gave a comparative account of the development of embryo-sac of eleven cucurbitaceous plants. Chakravorti (1947) reported the embryo development, normal tetrad formation and free nuclear type of endosperm in *Coccinia indica.* Singh (1955 *a*) studied the embryo development in *Cucumis melo.* Chopra and Basu (1965) recorded that the micropylar diad cell was much smaller than that of the chalazal end.

MATERIAL AND METHODS

The Kalianpur diploid plants of *Lagenaria siceraria* were raised in the garden of the Botany Section, Agricultural College, Poona. For observing pre-fertilization stages, female buds at anthesis and approximately 6, 12, 24, 48, 72 and 96 hours before anthesis were collected. In order to study the fertilization, the development of embryo, endosperm and seed, the ovaries of the pollinated flowers were collected at the interval of 3, 6, 9, 12 hours and 1, 2, 3, 4, 5, 6, 8, 10, 12, 14, 16, 18, 20, 23, 26 and 30 days after fertilization. In the advance post-fertilization stages the ovules were dissected from the previously fixed ovaries. The material was fixed in FPA (5 ml formalin + 5 ml propionic acid + 90 ml 70% alcohol) for the minimum period of 72 hours. The washing, dehydration, infiltration with paraffin and preparation of blocks, etc., were done as per the standard procedures. Sections were cut at the thickness of 8–12 μ for pre-fertilization stages and of 12–25 μ for post-fertilization stages. Haematoxylin staining technique was used.

RESULTS AND DISCUSSION

Five hundred and fifty six ovules at various growth stages were observed during the pre-fertilization study. The ovary was inferior with three carpels with parietal placentae which were bifurcated giving rise to six ovule bearing surface (Fig. 1). Development of ovules was not observed at the two extreme ends of the ovary. The ovules were found to originated from the placentae. The early indication of the development of an ovule was observed in the epidermal and a few sub-epidermal cells of each placenta towards the inner-side of the apex of the ovule initial (Fig. 2). The archesporium cut off parietal layers, the young megaspore mother cell got differentiated and an inner and outer integuments were formed (Fig. 3). The inner integument was composed of 2–3 layers of cells and outer integument was more massive and consisted of 7–8 cell layers (Fig. 4). This was in agreement with the description given by Chakravorti (1947), Mann and Robinson (1950) and Chopra and Basu (1965) who studied different species of the family cucurbitaceae.



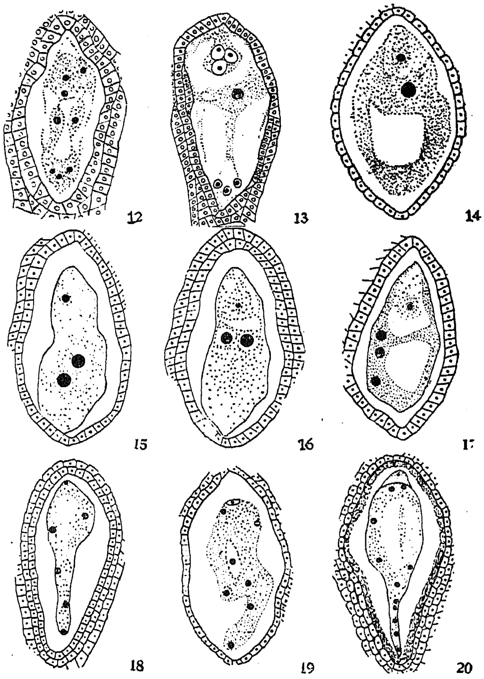
FIGS. 1–11

Immediately after the differentiation of ovule and development of integuments, the ovule was found to curve itself due to unequal growth rate and reached anatropous condition at anthesis. Chakravorti (1947) observed in *Coccinia indica* that a typical anatropous condition was attained after fertilization. The earliest differentiation of m.m.c. was observed below 1–2 cell layers occurred at 72 hours before anthesis. It was in agreement with the description given by Tillaman (1906) in *Cucumis sativus.* A primary sub-epidermal cell became differentiated into m.m.c. from other cells by its size and dark stained nucleus with hollow portion around it (Fig. 5). Megaspore mother cell pushed deep into fifth or sixth layer and after attaining the maximum size the nucleus of m.m.c. undergoes synizesis forming a diad (Fig. 6). The micropylar cell was smaller than chalazal one. At the end of second meiotic division a linear tetrad was formed (Fig. 7). Singh (1955 *a*) and Chopra and Basu (1965) also recorded similar observations in *Cucumis melo* and *Cucurbita* respectively. Out of four megaspores three of the spores at the micropylar end were found to degenerate and the cell at the chalazal end developed into embryo-sac mother cell (Fig. 8). The enlarged chalazal embryo-sac mother cell underwent three mitotic divisions and formed eight nucleated embryo-sac (Figs. 9, 10, 11 and 12). The mature embryo-sac was very small (58 μ × 30 μ) as compared to the size of the ovule (273 μ × 224 μ) inclusive of the inner and outer integuments. This study showed that in *L. siceraria* the development of female gametophyte conforms to the monosporic 8-nucleate polygonum type.

The eight nucleated embryo-sac was found to have two groups of four nuclei each at the two ends. One nucleus from each group moved towards centre constituting the two polar nuclei (Fig. 12). The two polar nuclei were first found to lie very close to each other but soon they fused and formed the secondary nucleus of the embryo-sac. Three nuclei at the chalazal end organized themselves into antipodal cells and three at the micropylar end gave rise to egg apparatus. The organization of mature embryo-sac therefore, consisted of two synergids, an egg, a secondary nucleus and three antipodal nuclei (Fig. 13). The antipodal nuclei were ephimeral in nature and degenerated before fertilization. Singh (1956 *a*) reported in *Citrullus vulgaris* that one or two antipodal cells persisted even after fertilization.

It was seen from Table I that m.m.c. was found to develop three days before the anthesis. Megasporogenesis was initiated two days before the initiation of anthesis. The diad and tetrad formation was observed mostly between 72 and 48 hours before anthesis. The mitotic divisions of embryo-sac mother cell took place 24 hours before and upto anthesis commonly. However, only about 12% of the ovules could reach the 8-nucleate stage at anthesis.

In the present investigations fertilization stages were not observed due to heavy staining of the embryo-sac. The pollen tube was observed in a few ovules at 48 hours after pollination. It traversed through style and entered the ovule porogamously. Porogamy appears to be the rule. Singh (1963 *a*) studied 22 cucurbits and observed that the pollen tube was porogamous. In most of the ovules fertilization was completed 72 hours after pollination.



FIGS. 12–20

For post-fertilization study 408 ovules were examined. It was seen from Table II that the megagametogenesis continued after anthesis. Soon after the fertilization the primary endosperm nucleus migrated towards the chalazal region and divided into two nuclei (Fig. 15) and this stage was found 5 days after pollination. Same results were obtained in *Coccinia indica* (Chakravorti, 1947) and *Cucumis melo* (Singh, 1955 *a*). The endosperm nuclei divided repeatedly and gave rise to free nuclei which took peripheral position in the embryo-sac and a vacuole was formed in the centre (Figs. 17 to 20). The endosperm, initially was free nuclear but in later stages it became cellular when the embryo was at 4-celled stage. Chopra (1955) observed similar results in *C. indica.* The endosperm was

TABLE I
Pre-fertilization stages of the diploid ovules

Sl. No.	Interval	Ovules observed	Stages					Vegetative ovules
			m.m.c.	mega-spore	nucleate embryo-sac			
					2	4	8	
1.	72 hours before anthesis	78	78	..	..	...	..	..
2.	48 hours before anthesis	120	37	78	..	..	..	10
3.	36 hours before anthesis	65	12	32	9	..	..	12
4.	24 hours before anthesis	138	..	46	39	7	—	45
5.	12 hours before anthesis	85	..	7	20	31	—	27
6.	At anthesis	70	..	..	6	30	8	24
	Total	556						

TABLE II
Post-fertilization growth stages of ovules

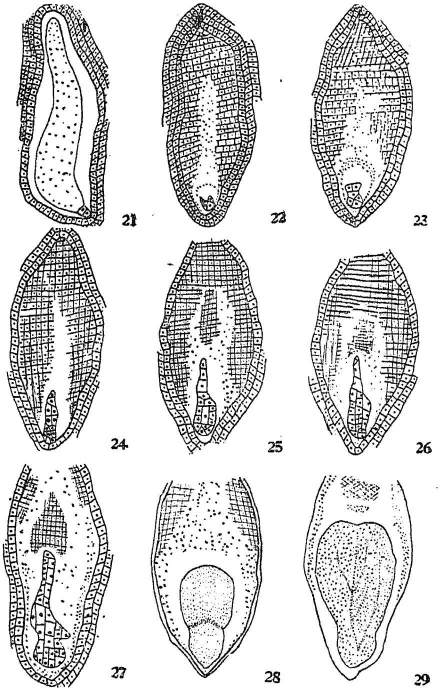
Sl. No.	Interval	No. of ovules	Embryo-sac with nuclei			Percentage of 8-nucleate embryo-sac
			2	4	8	
1.	3 hours	25	4	13	3	15
2.	6 hours	19	3	12	4	20
3.	9 hours	21	..	13	8	
4.	12 hours	30	1	14	10	40
5.	24 hours	28	11	14	13	47
6.	2 days	29	..	9	20	70
7.	3 days	24	..	2	22	93
			Embryo			Endosperm
8.	4 days	20	Zygote			1 nucleate
9.	6 days	15	Zygote			2 nucleate
10.	8 days	18	Zygote			4 nucleate
11.	12 days	24	Zygote			12 nucleate
12.	15 days	30	2-celled			78 nucleate
13.	18 days	25	4-celled			many and cellular
14	20 days	28	6-celled			do.
15.	23 days	20	12-celled			do.
16.	26 days	15	20-celled			do.
17.	30 days	12	30-celled			do.
18.	40 days	15	many celled · heart-shaped			do.
19.	45 days	10	many celled differentiated			do.
	Total	408				

differentiated into the haustorium at chalazal region (Fig. 18). The endosperm haustorium remained coenocytic and showed its maximum size (65 μ) when the embryo was heart-shaped. In *C. sativus* haustorium remained coenocytic (Chopra, 1953 *b*) but in *Citrullus colocynthis* it became cellular (Johri and Chowdhury, 1957).

The first division of zygote forming 2-celled pro-embryo was observed 15 days after pollination and when the endosperm reached 78-nucleate stage (Fig. 21). The cells of the pro-embryo divided transversely to give rise to 4, 6 and 12-celled pro-embryo (Figs. 22, 23 and 24) and the divisions in the pro-embryonal cells result in 16-celled, 20-celled, 30-celled pro-embryo (Figs. 25, 26 and 27) and finally a globular mass of embryo was formed. This globular embryo formed heart-shaped condition of the embryo (252 μ × 154 μ) at 40 days after pollination (Fig. 28).

A mature embryo with well developed cotyledons (700 μ × 350 μ) was found to occupy the seed cavity (Fig. 29) at 45 days after pollination.

Similar observations were recorded in *Bryonia dioica* (Soueges, 1939 *a*), *Citrullus colocynthis* and *Melothria maderaspatana* (Singh, 1961 *b*). The seed coat has its origin from the outer integument. The outer integument was constituted by a cell layer with prominent nuclei which later organized into seed coat. Similar results were obtained by Chakravorti (1947) in *Coccinia indica* and Singh (1953) in *Bryonia dioica, Luffa cylindrica* and *Cucurbita pepo.*



FIGS. 21–29

ACKNOWLEDGEMENTS

The author is grateful to Dr. R. D'Cruz, Ex Professor of Botany, College of Agriculture, Poona, and Dr. D. G. Hapse, Professor of Botany, College of Agriculture, Dhulia, for their keen guidance and kind encouragement.

1. Chakravarty, H. L., *Ind. J. Agric. Sci.,* 1946, 16, 1,
2. Chakravorti, A. K., *J. Indian Bot. Soc.,* 1947, 26, 95.
3. Chopra, R. N., *Proc. 40th Ind. Sci. Congr.,* 1953 *b,* p. 97.
4. —, *Phytomorphology,* 1955, 5, 219.
5. — and Basu, B., *Ibid.,* 1965, 15, 217.
6. Johri, B. M. and Chowdhury, C. R., *New Phytol.,* 1957, 56, 51.
7. Kirkwood, J. E., *Bul. N.Y. Bot. Gard.,* 1905, 3, 313.
8. Mann, L. K. and Robinson, J., *Amer. J. Bot.,* 1950, 37, 685.
9. Singh, B., *Phytomorphology,* 1953, 3, 224.
10. Singh, D., *J. Indian Bot. Soc.,* 1955 *a,* 34, 72.
11. —, *Proc. 43rd Ind. Sci. Congr.,* 1956 *a,* 3, 222.
12. —, *J. Indian Bot. Soc.,* 1961 *b,* 40, 620.
13. —, *Ibid.,* 1963 *a,* 42, 208.
14. Soueges, R., *Acad. Sci. Paris,* 1939 *a,* 208, 227.
15. Tillaman, O. J., *Ohio Nat.,* 1906, 6, 423.

THE INDIAN NATIONAL SCIENCE ACADEMY, NEW DELHI-1, AWARDS

The Silver Jubilee Commemoration Medal, for 1976, was awarded to Prof. A. K. Sharma, FNA, in recognition of his contributions in the field of Cytogenetics and Cytochemistry.

The Shree Dhanwantari Medal was awarded to Dr. B. Mukerji, FNA, in recognition of his contributions to medical and drug research in India.

The Professor T. R. Seshadri Seventieth Birthday Commemoration Medal (1976) was awarded to Prof. R. C. Mehrotra, FNA, Vice-Chancellor, University of Delhi, for his contributions on the Chemistry of Coordination Compounds.

LETTERS TO THE EDITOR

DIELECTRIC INVESTIGATION OF RELAXATION IN SOME POLAR MOLECULES

THE present communication reports the experimental as well as calculated values of relaxation times of 2-4-6-tribromoaniline, *o*-, *m*-, *p*-dimethoxy benzenes; *p*-diethoxy benzene, 2-nitroanisole and 4-nitroanisole in 3 cm microwave region at 20° C in dilute solutions.

It has been observed that Debye[1] equation and its several modifications (Perin[2], Hill[3] and Meakins[4]) yielded results which are not in good agreement with the observed values. It is therefore planned to calculate the relaxation time using Wirtz and Coworkers[5] equation.

The dielectric constant (ϵ') and dielectric loss factor (ϵ'') have been determined using the standing wave technique of Roberts and VonHippel[6] and its subsequent simplification by Dakin and Works[7]. The relaxation time was then determined using the concentration variation method of Gopala Krishna[8] discussed in an earlier paper[9].

The experimental values of relaxation time along with those calculated using Wirtz and Coworkers'[5] equation are given in Table I.

TABLE I

Compound	τ exp × 10^{12} sec	τ Wirtz and Coworkers × 10^{12} sec.
2-4-6 tribromo aniline	14·11	21·48
o-dimethoxy benzene	10·79	18·42
m-dimethoxy benzene	9·96	17·69
p-dimethoxy benzene	5·39	19·52
p-diethoxy benzene	5·91	18·26
2-nitroanisole	15·71	17·91
4-nitroanisole	9·45	18·24

The relaxation times of *o*- *m*- and *p*- dimethxoy benzenes decrease from ortho *via* meta to para compounds which indicates that the intramolecular rotation of the methoxy group in ortho dimethoxy benzene experiences a greater hindrance as compared to the meta and para dimethoxy benzenes. *p*-diethoxy benezene exhibits a slightly higher relaxation time than *p*-dimethoxy benzene. This means under similar conditions methoxy group has greater freedom of rotation than ethoxy group.

The relaxation time calculated using Writz and Spernol equation gives better agreement with experimental values, than those derived using Debye equation (using η and $0 \cdot 36\,\eta$). It is therefore concluded that Wirtz and Coworkers equation is a better representation of phenomenon of relaxation.

The Authors are deeply indebted to Prof. B. G. Gokhale, for his interest and encouragement and Dr. M. C. Saxena for his guidance.

Physics Department, N. K. MEHROTRA.
Lucknow University, S. K. S. SOMEVANSHI.
Locknow (U.P.), *November* 25, 1975.

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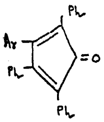
1. Debye, P., *Polar Molecules*, Reinhold Publishing Corporation, New York, 1929, Chapt. 5.
2. Perin, J., *J. of Phys. Radium*, 1934, 5, 497.
3. Hill, H. E., *Proc. of Physical Society*, (Lond.), 1954, 67 B, 149.
4. Meakins, R. J., *Trans. Faraday Society*, 1958, 54, 1160.
5. Wirtz, K. and Spernol, A., *Z. Naturforsch.*, 1953, **8 A**, 522.
6. Roberts, S. and Von Hippel, A., *J. of Appl. Phys.*, 1946, 17, 610.
7. Dakin, T. W. and Works, C. N., *Ibid.*, 1947, 18, 789.
8. Gopala Krishna, K. V., *Trans. Faraday Society*, 1957, 53, 767.
9. Mehrotra, N. K., Shukla, J. P. and Saxena, M. C., *Indian J. Pure Appl. Phys.*, 1967, 5, 61.

USE OF TETRACYCLONE DERIVATIVE IN THE SYNTHESIS OF POLYPHENYLATED AROMATICH YDROCARBONS BY DIELS-ALDER REACTION

TETRACYCLONE, (2, 3, 4, 5-tetra phenyl cyclopentadienone), as diene, has been successfully used by many workers for the synthesis of polyphenylated aromatic hydrocarbons through Diels–Alder reaction[1-5]. Reactions of derivatives of tetracyclone, in which one or more phenyl groups carry monosubstitution have also been reported[6-10]. However, tetracyclone with one or more disubstituted phenyl groups and their reactions are not well known. In this communication we report the synthesis of three isomers of 3-xylyl-2, 4, 5-triphenyl cyclopentadienone (I) and their reactions with several dienophiles, which include maleic anhydride, bicycloheptadiene, diphenyl acetylene, benzyne and acenaphthylene. Thus, several types of polyphenylated aromatic hydrocarbons have been synthesised. Three isomeric cyclopentadienones (I) have been obtained from condensation of the corresponding isomeric dimethyl benzils, (*viz.*, 3, 4-, 2, 4-, and 2, 5-dimethyl benzils) and dibenzyl ketone using Triton B as catalyst (yield, 85%) instead of usual ethanolic alkali. The latter catalyst gave very poor yields (10%) particularly in the case of dimethyl benzils, containing methyl group in ortho position with respect to C = O group, probably due to benzilic acid type rearrangement[11]. The dimethyl benzils have been prepared by oxidation of the corresponding deoxybenzoins with selenium dioxide-acetic anhydride[6].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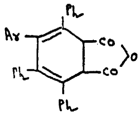
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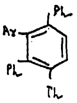
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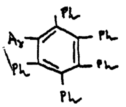
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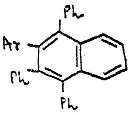
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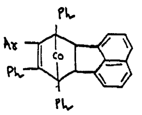
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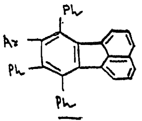
VII

where Ar = a) = o-$(CH_3)_2C_6H_3$-
b) = m-$(CH_3)_2C_6H_3$-
c) = p-$(CH_3)_2C_6H_3$-

Maleic anhydride reacts with I when refluxed in bromobenzene to form adducts (II) which when distilled with soda-lime undergo decarboxylation and thermal dehydrogenation to give three isomeric dimethyl-2′, 6′,-diphenyl-*m*-terphenyl (III). The same aromatic hydrocarbons are obtained directly on refluxing I and bicycloheptadiene in chlorobenzene medium with the loss of carbon monoxide and cyclopentadiene. Diphenyl acetylene undergoes fairly rapid reaction with I in benzophenone medium to yield three isomeric dimethyl-2′, 4′, 5′, 6′-tetraphenyl-*m*-terphenyl (IV). Bezene, produced *in situ* by aprotic diazotisation of anthranilic acid[12], when trapped with I yields 1, 2, 4-triphenyl-3-xylyl-naphthalenes (V). Use of acenaphthylene as dienophile leads to fluoranthene derivatives and this reaction is carried out in a low boiling solvent, *e.g.*, benzene makes isolation of adducts (VI) possible whereas tetralin, a high boiling solvent leads directly to 7, 8, 10-triphenly-9-xylyl-fluoranthenes (VII).

All compounds are crystallised from benzene-alcohol and all are colorless except I and VII, the former being black and the latter yellow. Melting points shown in Table I are uncorrected and satisfactory analytical (C and H) results of the compounds have been obtained. The ultraviolet absorption spectra of the three isomers of I (Table II) in benzene show two absorption maxima in each case. The absorption maxima in the lower wavelength region have been attributed to C = C absorption and that in higher wavelength to C = O absorption[13]. The infra-red spectral studies on three isomeric tetracyclones (I) show a prominent carbonyl band at 5·85 μ. I.R. and N.M.R. spectra of several of these aromatic hydrocarbons (III to V and VII) have been measured and subsequently included in the "Standard Spectra" by Sadtler Research Laboratories, Philadelphia, U.S.A.

TABLE I
Melting points (° C)

	(*a*)	(*b*)	(*c*)
I	162	167	152
II	203	196	176
III	125	145	156
IV	>340	>340	>340
V	147	213	138
VI	203 (decomp.)	191 (decomp.)	196 (decomp.)
VII	234	203	221

TABLE II
U.V. absorption of 3-*xylyl*-2, 4, 5 *triphenyl cyclopentadienone*

	$\frac{I_{max}}{\lambda_{nm}}$ (ϵ)	$\frac{II_{max}}{\lambda_{mn}}$ (ϵ)
I (*a*)	355 (7100)	518 (1233)
I (*b*)	345 (6333)	518 (1566)
I (*c*)	345 (6250)	518 (1633)

The authors wish to thank Dr. S. Thakur, of Department of Chemistry, University of Burdwan and Prof. A. De, of S. B. College, Magra, Hooghly, for their deep interest in the work.

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1. Alder, K., *New Methods of Preparative Organic Chemistry*, Interscience Publishers, Inc., New York, 1948, p. 381.

2. Norton, J. A., *Chem. Rev.*, 1942, 31, 319.
3. Pelaez, R. M., *Afinidad,* 1953, 30, 133.
4. Dilthey, W., *Chem. Zentr.*, 1936, 1071, 2838.
5. Allen, C. F. H. and Sheps, L. J., *J. Res.*, 1933, 8, 414.
6. Coan, S. B., Trucker, D. E. and Becker, E. I., *J. Amer. Chem. Soc.*, 1953, 75, 900.
7. Mehr, L., Becker, E. I. and Sooerri, P. E., *Ibid.*, 1934, 141, 331.
8. Dilthey, W., Torsken, O., Plum, K. and Schomer, W., *J. Prakt. Chem.*, 1934, p. 141.
9. Friedman, A., Gugig, W., Mehr, L. and Becker, E. I., *J. Org. Chem.*, 1959, 24, 526.
10. Coan, S. B., Trucker, D. E. and Becker, E. I., *J. Amer. Chem. Soc.*, 1955, 77, 60.
11. Shapiro, E. L. and Becker, E. I., *Ibid.*, 1953, 75, 4769.
12. Logullo, F. M., *Ibid.*, 1963, 85, 1549.
13. Berman, E. D. and Hirshberg, Y., *Bull. Soc. Chim.*, France, 1950, 1091, 17.

PYRIDINE-2-ALDOXIME AS A REAGENT FOR THE SPECTROPHOTOMETRIC DETERMINATION OF RUTHENIUM (III) IN AQUEOUS MEDIUM

Pyridine-2-aldoxime has already been used for the determination of transition metals, platinum and palladium[1-3]. As a result of the investigations on pyridine-2-aldoxime complexes with platinum metals, it was observed that on boiling ruthenium (III) solutions (taken as chlororuthenate) for 2–3 min., a dark brown water soluble complex is gradually formed which can be used for the spectrophotometric determination of ruthenium. As ruthenium (IV) combines more readily with pyridine-2-aldoxime in the presence of reducing agents (hydroxylamine or sulphite), it is believed that the reddish brown complex contains ruthenium (III).

The optimum conditions for the spectrophotometric determination of ruthenium (III) with pyridine-2 aldoxime are given in the following *recommended-procedure.*

Recommended Procedure.—To 1 ml of ruthenium (III) solution in acid solutions (1 N) and upto a pH of 4·5, and containing 125–500 μg Ru for 1 cm path length, is added 5 ml of 0·05% w/v aqueous soltion of pyridine-2-aldoxime and the solution boiled for about 5 min. On cooling and diluting to 25 ml with water, the absorbance of the solution is measured at 420–430 nm using water as blank. The concentration of ruthenium is found out from a calibration curve prepared under similar conditions. The relative standard deviation of the method in the recommended range is worked out to be ± 1·0%.

Characteristics of the Reddish Brown Complex

The absorption spectra of the reddish brown ruthenium-pyridine-2-aldoxime complex shows the presence of only a single absorption maximum at a wavelength 420–430 nm. After boiling the solutions and developing the colour fully, the absorbance at 425 nm remains constant over the pH range 1·6–11·3. However, before boiling, ruthenium solutions should be acidic (1 N) and may be upto a pH 4·5, otherwise the colour development is incomplete. Minimum stoichiometric concentration ratio of ruthenium to pyridine-2-aldoxime for full colour development under these conditions is 1:8. Molar absorption of the reddish brown complex is found out to be $2 \cdot 34 \times 10^3$ l. cm^{-1}· mole^{-1} (420 nm). The concentration range of ruthenium (III) for which Beer's law is obeyed, is found to be 3·0–3·50 μg/ml.

The stoichiometric ratio of ruthenium (III) to pyridine-2-aldoxime in the complex is found to be 1:2 by the continuous variations method. Because pyridine-2-aldoxime is a tautomeric molecule having potentiality of bonding to platinum ion in either syn (N + N) or anti (N + O) form [4,5], the stability constant of the complex in the solution was not determined. However, the conditional formation constant under these conditions was determined to be as $\log k = 3 \cdot 40 \pm 0 \cdot 10$.

Interferences due to Foreign Ions

For the purpose of the determination of interferences, a ± 2% deviation from the expected absorbance was the criterion. It was observed that for the determination of ruthenium (III), even 100-fold concentrations (by weight of bromide, iodide, sulphate, oxalate, nitrate, tartrate, citrate, magnesium, calcium, barium, strontium, mercury, zinc and cadmium do not cause any interference. Maximum amounts of other ions that can be tolerated for 50 μg Ru are: 15 μg rhodium (III), 30 μg irridium (III), 20 μg osmium (IV), 2 μg platinum (IV), 50 μg gold (III), 200 mg iron (II), 100 μg cobalt (II) and 70 μg nickel (II). Lead (II) and silver are precipitated as chlorides in HCl media, whereas palladium (II) quantitatively precipitated by pyridine-2-aldoxime, hence centrifugation before absorbance measurements eliminates the interferences due to these ions.

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1. Krause, R. A., *Dissert. Abstr.*, 1960, **20**, 3504.
2. Kumar, N., *Ph.D. Thesis*, University of Delhi, 1973.
3. Liu, C. F. and Liu, C. H., *J. Amer. Chem. Soc.*, 1961, **83**, 2615; *Inorg. Chem.*, 1964, **3**, 678; 1963, **2**, 706.
4. Hanania, G. I. H. and Irvine, D. H., *Nature*, 1959, **183**, 40.
5. Blackmore, M. W. and Magee, R. J., *J. Inorg. Nucl. Chem.*, 1969, **31**, 2859.
6. Narayan Kumar, Manku, G. S., Bhat, A. N. and Jain, B. D., *Talanta*, 1970, **17**, 873.

EFFECT OF ISATIN ON CHICK BRAIN ACID AND ALKALINE PHOSPHATASES

In brain several vitamin-B_6 dependent enzymes influence the level of γ-amino butyric acid (GABA)[1] whose deficiency has been implicated as a possible causative factor of convulsive seizures.[2] Since Turner has postulated that phosphatases may be involved in the regulation of vitamin-B_6 requiring enzymes[3], it was thought desirable to investigate the effect of isatin, a promissing anticonvulsant agent[4,5] on brain phosphatases.

Male adult white-leg horn chicks were used for these studies. For *in vivo* experiments, twelve chicks were divided into two groups of six each. The animals of the first group served as control while those of the second group were fed with isatin in a single dose of 200 mg/kg body weight, the active dose of isatin for its anticonvulsant action in electroshock seizures[4]. The chicks of the second group were killed by decapitation without anaesthesia after two hours of isatin administration, the peak period of its action[4]. The different parts of the brain (cerebellum, cerebral hemisphere and optical lobes) were removed and homogenized with ice cold water. The supernatants obtained after centrifugation of these homogenates were used as the source of enzyme. Acid and alkaline phosphatase activities were assayed by the method of Kind and King[6] using phenyl phosphate as substrate and citrate buffer (100 mM) of pH 4·9 and carbonate-bicarbonate buffer (100 mM of pH 9·6 respectively at 37°.

In the *in vitro* experiments acid and alkaline phosphatase activities of adult chick brain were moderately inhibited by 32·4 and 44·7%, respectively by 10 mM isatin. However, *in vivo* administration of this compound evoked opposite response in acid and alkaline phosphatases. After two hours of isatin administration, the acid phosphatase activity showed an increase of 23·0%, 33·3% and 16·5% in cerebellum, cerebral and optical lobes respectively. Although the increases are not very large, statistically these are highly significant (Table I). No significant change in the alkaline phosphatase activities of cerebral hemisphere and optical lobes but a slight statistically significant lowering (15%) in the activity of cerebellum enzyme was observed.

TABLE I

Effect of isatin on acid and alkaline phosphatase activities of different parts of chick brain 2 hours after its administration

	Enzyme Activity		% increase	P value
	without drug	with drug		
I *Acid phosphatase (mg phenol/hr/g of fresh tissue)*				
Cerebellum	4·24±0·07	5·32±0·10	25·4	<0·01
Cerebral hemisphere	4·86±0·04	6·48±0·07	33·3	<0·01
Optical lobes	4·41±0·04	5·14±0·06	16·5	<0·01
II *Alkaline phosphatase (mg phenol/15 min/g of fresh tissue)*				
Cerebellum	2·38±0·06	2·02±0·08	15·0	<0·01
Cerebral hemisphere	1·03±0·03	1·00±0·04	..	N.S.
Optical lobes	4·11±0·06	4·10±0·08	..	N.S.

N.S. : Not significant.

Roberts *et al.*[7] as well as Begum and Bachhawat[8] have demonstrated the presence of pyridoxal phosphate phosphatase. They found this enzyme to be an acid phosphatase with broad specificity. An increase in the activity of this enzyme may lead to deficiency of pyridoxal phosphate which in turn leads to deficiency of GABA and hence convulsions[2]. In the present study, although isatin administration produces an increase in acid phosphatase activity, yet it does not cause convulsions (rather it prevents them) showing that this increase is not associated with pyridoxal phosphate phosphatase. At present we are unable to fathom the full implication of the increase of acid phosphatase activity by isatin.

In vitro inhibition and *in vivo* activation of the acid phosphatase by isatin may be explained on the basis of its reported breakdown in the body[9].

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1. Harper, H. A., *Review of Physiological Chemistry*, 13th Edition. Kothar Book Depot, Bombay, 1971, p. 347.

2. Narang, B. S., Talwar, G. P. and Singh Baldev, *Ind. J. Physiol. Pharmacol.*, 1962, **6**, 1.
3. Turner, J. M., *Biochem. J.*, 1960, **80**, 663.
4. Sareen, K. N., Kohli, R. P., Amma, M. K. P. and Gujral, M. L., *Ind. J. Physiol. Pharmacol.*, 1962, **6**, 87.
5. Mueller, M., *Med. Exptl.*, 1962, **7**, 155.
6. Kind, P. R. N. and King, E. J., *J. Chim. Pathol.*, 1954, **7**, 322.
7. Roberts, E., Rothstein, M. and Baxter, C. F., *Proc. Soc. Exp. Biol. N.Y.*, 1958, **97**, 796.
8. Begum, A. and Bachhawat, B. K., *Ann. Biochem.*, 1960, **20**, 143.
9. Sakameto, Y., Kematani, K., Inoue, F. and Kotake, M., *Symposia on Enzyme Chemistry* (Japan), 1956, **11**, 242.

A REVERSIBLE SPERMIOSTATIC FACTOR PRESENT IN BUFFALO SEMINAL PLASMA

THE occurrence of initially non-motile but revivable spermatozoa in occasional buffalo semen samples was reported earlier[1]. The preliminary findings on the possible basis of this peculiar phenomenon of buffalo semen are briefly presented here. pH, tonicity, fructose level and cation distribution (Na, K and Ca) in the seminal plasma did not appear to have any direct implication in the occurrence of this phenomenon since the non-motile and motile ejaculates were indistinguishable in respect of these variables. It was found that a factor located in the seminal plasma was responsible for the reversible inhibition of spermatozoan motility since washed motile spermatozoa from normal ejaculates were found to be reversibly immobilised when resuspended in cell free seminal plasma obtained from non-motile ejaculates and *vice versa*. On fractionation of the seminal plasma from non-motile ejaculates by dialysis in a cellophane tube at 2° C against three changes of distilled water every 48 h and redialysis of the non-dialysable portion against isosmotic phosphate buffer (pH 7·4), it was found that the motility-inhibitory factor was located in the non-dialysable fraction. The factor was fairly heat-stable (90° C for 1 min.) and possibly, therefore, non-enzymic in nature.

In the presence of cysteine, glycine and citrate-which share the common property of chelating heavy metals (Cu, Zn, etc.), the inhibition of motility in revivable non-motile ejaculates was found to be instantaneously released indicating the reversible nature of the phenomenon. Addition of glucose or fructose did not appear to erevice the motility of non-motile ejaculates. It is difficult to say if the motlity-inhibtory factor of buffalo seminal plasma and the so-called toxic factor claimed to be present in bull seminal plasma adversely affecting sperm viability during *in-vitro* storage[2] are chemically identical, though there are a few points of apparent similarities between the two. The instantaneous revival of motility in initially non-motile ejaculates by chelating agents is suggestive that the inhibition may be operative at the level of ATP-ase enzyme which is involved in the enzymic breakdown of ATP providing immediate energy for sperm movement. It is conceivable that the motility-inhibitory factor may, in fact, be an organo-metallic compound which acts as a potent ATP-ase inhibitor and is neutralised in the presence of metal-binding agents. Further work on chemical characterization of the factor is in progress.

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1. Abbhi, H. L., Sengupta, B. P., Roy, A. and Sahani, K. L., *Indian J. of Vet. Sci.*, 1968, **38**, 253.
2. Shannon, P., *J. Dairy Sci.*, 1965, **48**, 1362.

PROTECTIVE EFFECT OF CHLORPROMAZINE ON TISSUE CULTURE AGAINST POLIOVIRUS

MANY antihistaminic drugs protect tissues against damaging effects of viruses due to stabilization of the lysosomal membranes[1-3]. Damage to different subcellular organelles of tissue occurs during poliovirus infection[4]. The present experiments demonstrate that chlorpromazine stabilizes membranes of different subcellular organelles besides lysosomes, of monkey kidney tissue culture and consequently protects it against poliovirus infection.

Monkey kidney tissue cultures were prepared in bottles[5]. To one group of culture bottles, poliovirus type I was inoculated in doses of 1 ml of about 100 TCID$_{50}$ per 0·1 ml. In the second group chlorpromazine (Largactil—May and Baker) was mixed with the tissue culture fluid so that its concentration was 10^{-5} M. The third group had poliovirus and chlorpromazine while the fourth group was used as untreated control·

All the bottles were examined for morphological changes after incubating at 37° C for 24 hours when the experiment was terminated. The tissue culture cells were prepared and acid and alkaline phosphatase and adenosine triphosphatase were estimated as described before[5,6]. For the estimation of 5′-nucleotidase, the test reaction mixture consisted of 1·0 ml tris buffer (pH 7·5) and 0·1 ml of manganese sulfate. The control reaction mixture consisted of 1 ml of tris buffer (pH 7·5) and 0·1 ml manganese sulfate and 0·2 ml of nickel chloride. To both tubes, 0·2 ml of cell homogenate was added and these were placed in a water bath at 37° C for 3 minutes. Then 0·2 ml of adenosine-5′-phosphate was added and after 30 minutes the reaction was stopped by the addition of 2 ml of 10 per cent trichloracetic acid. The tubes were centrifuged for 5 minutes and suitable aliquots were taken to determine inorganic phosphorus[7]. The details of the reagents, etc., have been reported by Gupta *et al.*[8].

It was observed in the present study that poliovirus type I produced a distinct cytopathogenic effect in the monkey kidney tissue cultures after 24 hours. In contrast, poliovirus failed to produce any cytopathogenic effect in tissue cultures treated with chlorpromazine. It was further observed that all the four enzymes, *viz.*, acid and alkaline phosphatase, 5′-nucleotidase and adenosine triphosphatase were minimal in tissue cultures treated with chlorpromazine. The values of the enzymes were maximal in the cells treated with the poliovirus alone. But the increase in the enzyme contents by the poliovirus was reduced if the cells were treated with chlorpromazine (Table 1).

TABLE I

Enzyme contents in tissue culture cells in different groups

Groups	ENZYMES			
	Acid phosphatase KAU	Alkaline phosphatase KAU	5′-nucleotidase mg P/100 ml	Adenosine triphosphatase P/100 ml.
Group I Poliovirus	1·06	1·66	7·26	2·42
Group II Chlorpromazine	0·17	0·36	4·85	0·82
Group III Poliovirus and Chlorpromazine	0·73	0·55	5·85	1·00
Group IV Control	0·60	0·40	7·25	1·04

Control—untreated tissue cultures.

Eyal *e al.*[9] and Alho[10] have shown that chlorpromazine protects lysosomes. However, our findings show that chlorpromazine, besides stabilizing the lysosomal membrane (acid phosphatase), also stabilizes the mitochondrial (5′-nucleotidase and adenosine tri-phosphatase), and the plasma membranes (alkaline phosphatase) and protects them from the damaging effects of the poliovirus as shown by decrease in the maker enzymes of the respective subcellular organelles.

We had observed in the pilot experiments of this study that chlorpromazine does not affect the infectivity of the poliovirus. Therefore, inactivation of the virus does not appear to be the cause of protection of tissue cutlture. This is similar to what has been reported for antihistaminic drugs[3]. Protection of the cells by chlorpromazine may thus be brought about by preventing damage to essential enzyme systems of cells (*e.g*, mitochondrial and plasma membrane enzymes) and by inhibiting the release of hydrolytic lysosomal enzymes by the virus.

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---

1. Judah, J. D., Bjovedt, G. and Vianio, D., *Nature*, 1960, **187**, 507.
2. Vainio, T., Judah, J. D. and Bjovedt, G., *Exp. Mol. Path.*, 1962, **1**, 15.
3. Piplani, S. K. and Aikat, B. K., *Indian J. Path. Bact.*, 1971, **14**, 68.
4. Chaturvedi, U. C., Bahuguna, L. M. and Mehrotra, R. M. L., *Indian J. Med. Res.*, 1972, **60**, 589.
5. Bahuguna, L. M. and Chaturvedi, U. C., *Brit. J. Exp. Path.* 1974, **55**, 314.
6. Chaturvedi, U. C., Agarwal, D. K., Bahuguna, L. M., Mathur, A. and Mehrotra, R. M. I., *Curr. Sci.*, 1975, **44**, 774.
7. Fiske, C. M. and Subba Row, V., *J. Biol. Chem.*, 1925, **66**, 375.
8. Gupta, M. B., Bhalla, T. N., Tangri, K. K. and Bhargava, K. P., *Biochem. Pharmacol.*, 1971, **20**, 401.
9. Eyal, Z., Warwick, W. J., Mayo, II, C. H. and Lillehei, R. C., *Science*, 1965, **148**, 1468.
10. Alho, A., *Biochem. Pharmacol.*, 1973, **22/20**, 2521.

RADIOMETRIC DATING OF A LITHIUM PEGMATITE NEAR AJMER

Introduction

THE paper pertains to the determination of geological age of the pegmatites occurring at the village Rajagarh near Ajmer. The determinations are done on lithium minerals namely (Rubellite and Lepidolite), by an uncorrected total Sr method, using Atomic absorption Spectrometer and Neutron activation analysis). The ages determined are in close agreement with the ages given by Vinogradova[1] (1966) and Craford[2] (1970) by the methods of potassium-argon and Rb/Sr, using Mass Spectrometer.

Geological Setting

Lithium bearing pegmatites occur in a quarry near the village Rajagarh about six miles northeast of Ajmer town. The country rocks are mostly biotite-quartz schists and biotite gneisses lying in broad folds, probably parallel to the trend of the Aravali belt of rocks. These country rocks have been intruded by granites and pegmatites into which lithium was later introduced.

The pegmatite is concordant with the country rocks and the whole pegmatite has a form, which is somewhat elongated and tabular with a definite hanging wall of the country rock. The pegmatite shows different zones characterised by different mineral composition and texture. The upper portion is different from the lower part in mineral composition as well as texture. The middle portion is characterised by lithium mineralisation.

Field and megascopic study reveal three stages of mineralisation. Before folding, the gneisses were invaded by solutions which resulted in lit-par-lit injection of quartz and felspar pegmatite with black tourmaline. In numerous instances, the felspar individuals or the pegmatite are granulated or drawn out into augen shapes. After the lit-par-lit injection, there were two periods of pegmatite formation. The two stages of pegmatites are characterised by lithium tourmaline (rubellite) and lithium mica (lepidolite) mineralisation. The second stage pegmatite is characterised by rubellite, apatite and beryl ; while the third stage is characterised by lithium mica, namely, lepidolite and sporadic zinnwaldite. The mineralogy of rubellite and lepidolite has been worked by the author in earlier publications (Babu[3,4]).

Method of Study

The minerals rubellite and lepidolite were hand picked by chistling it from the outcrops. They were crushed to pass through 80 mesh. washed and dried. This material was subjected to heavy liquid separation (Clerici's solution with various dilutions) to obtain a pure crop of the minerals. The material separated was almost pure. This material was further ground to fine powder and one gram of the ground material was accurately weighed and dissolved in A.R. perchloric acid and HF, and extracted with A.R.HCl. This solution was used for the determination of strontium and rubidium for age determination of the pegmatites.

Results

(*i*) *Age determination using Rubellite.*—A gross radiometric age (1294 ± 200 m.y) uncorrected for non-radiogenic strontium, for rubellite is computed from rubidium (100 ppm, determined by atomic absorption spectrometer) and strontium (50 ppm, determined by atomic absorption spectrometer) contents ($Rb^{87} = 1 \cdot 39 \times 10^{11}$ years). As rubellite has been formed in the pegmatite by replacement, subsequent to the formation of post-Aravali pegmatites of Ajmer, the gross radiometric age does not depict the actual age of the pegmatite. However, the gross radiometric age determined on rubellite is in fair agreement with, with the Rb/Sr age of 1600–1700 m.y. given by Crawford[2] (1970) for the pegmatite of Rajasthan.

(*ii*) *Age determination using Lepidolite.*—A gross radiometric age has been computed from the lepidolite in the following manner :

Total rubidium content = 2470 ppm (determined by Atomic absorption spectrometer and Neutron activation analysis)

$$\text{Rubidium}^{87} \text{ content} = 2470 \times 0 \cdot 278 = 678 \text{ ppm}$$

Total strontium content = 100 ppm (determined by Atomic absorption spectrometer)

$$\text{Strontium}^{87} \text{ content} = 100 \times 0 \cdot 0702 = 7 \text{ ppm}$$

(assuming the abundance of normal strontium)

$$Rb^{87} = 1 \cdot 39 \times 10^{-11} \text{ years}$$

$$\text{Gross radiometric age of the mineral} = \frac{1}{1 \cdot 39 \times 10^{-11}} \times \frac{7}{687}$$

$$= 733 \pm 150 \text{ m.y.}$$

(assuming that all strontium in the mineral is radiogenic)

The gross radiometric age gives the probable age of the metamorphic-metasomatic episode, but not actual age of the pegmatite in which the mineral occurs. This probable age computed for the mineral lepidolite is in close agreement with the potassium-argon age given for the lepidolite from the pegmatite of Rajagarh by Vinogradova[1] (1966, 913 m.y).

The studies instituted agrees well with the field observation of the two stages of pegmatites in which the pegmatite containing rubellite is earlier and the pegmatite containg lepidolite is later (Age of rubellite pegmatite 1294 ± 220 m.y ; Age of lepidolite pegmatite 733 ± 150 m.y). Besides, these age determinations closely agree with the age determinations by mass spectrometry published by Vinogradova and Craford, within the permissible limits of instrument and technical errors.

The author thanks Dr. U. Aswathanarayana, for his helpful suggestions in preparing this paper. My thanks are also due to Heads of the Divisions of Analytical Chemistry and Health Physics of "BARC," Bombay.

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Sagar, M.P., *July* 3, 1975.

---

1. Vinogradova, A., *Geochronologia Dokendriya Indii*, Tr. Akad Nauk, U.S.S.R., Komopredelariyu Absolyut, Vozrasta, Geol. Forma, 1966, 13, 394 (Russian Translation).
2. Crawford, A. R., *Can. Journ. Earth. Sci.*, 1970, 7 (1), 91.
3. Babu, S. K., *Journ. Geol. Soc. Sagar*, 1970, 6, 1.
4. —, *Journ. Univ. Sagar*, 1970, 18 (18), 11.

A NOTE ON THE CONTROL OF JOINTING BY TOPOGRAPHY BETWEEN JANGAL GALI AND SALAL, JAMMU PROVINCE, J AND K STATE, INDIA

Introduction

THE present note seeks to analyse for the first time the concept of control of jointing by topography in an area between Jangal Gali (Lat. 33° 01′ 15″ N: Long 75° 01′ E) and Salal (Lat. 33° 10′ N: Long. 74° 50′ E) in the Udhampur District of Jammu. The area has a variety of landforms ranging from high mountain ranges on the north to flat plains on the south. The ruggedness of the topography increases inland on the north where limestones become dominant. The hogback shaped masses of limestones range in altitude from 600 to 2700 meters above the mean sea level. Throughout the terrain, the structure, is reflected by the dendritic pattern of the streams of all the orders, the Chander Bhagga or the Chenab is the only consequent stream of importance in the area.

The "Great Limestone" is the oldest folded for mation whose base has not been seen anywhere in the whole terrain. On the basis of some stromatolitic bands found (Chadha; 1975) associated with the formation, an upper pre-cambrian age has been ascribed to it. Among the early workers mention may however be made of Medlicott[5] (1864), Wadia[8] (1928), Gundu Rao[7] (1970), and Raha and Sastry[4] (1973). None of the early workers has explained the interdependence of jointing and topography.

Nature of Jointing

Conventionally, joints are defined as fully developed fractures in rocks along which there has been no relative displacement. The joints show varying degree of dilations. The incipient and tightly closed joints are generally irregular, more in number and are represented by cleaved grains. Such joints are mostly due to deformative forces and decrease in number with depth. True joints have great dilations and are easily recognised. The dilation of joints depends mostly on crustal deformations. The sliding under the force of gravity favours the opening of steep incipient fractures and as such the trends of the joints are controlled mostly by the slide directions.

In the Jangal Gali the general topography is mountainous and rugged with transverse and oblique streams which have formed deep valleys and gorges with steep escarpments. The "Great Limestone[44] formation is flanked by a thick pile of Murrees and still younger Tertiaries. The constant sliding of the Murrees produces incipient fractures. The important sets of joints observed in this terrain are:—

(*a*) N 45° E–S 45° W, dipping 30° towards S 36° E.
(*b*) N 25° E–S 25° E, dipping 55° towards S 75° E

Most of the joints (Figs. 1 and 2) are striking parallel to the beds, but dipping across bedding and some of them are striking at an angle to the bedding but dipping 45–50° towards SE. Of the many vertical joints, one set is in the strike direction while the other in N 20° E–S 20° W direction. The strike of joints is relative to the topography.

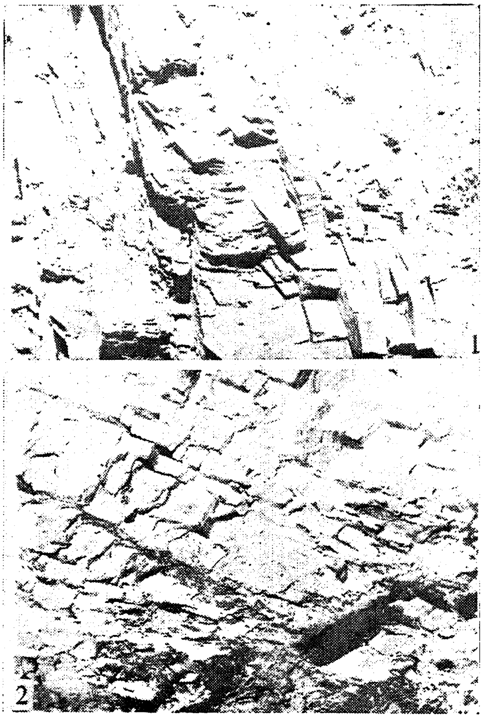
At Katra (Lat. 32° 59′ 30″N: long 74° 56′ E) which has rugged topography, some of the peaks such as Trikuta (2700 meters) are the highest and remain under snow for most part of the year. The Balganga is an important stream of the area. The hogback topography of the area has revealed a net work of simple as well as intricate joints. Most of the topographical joints have been deepened by solution work. Such steep and narrow joints have been designated as CLINTS or GRYKES in England, LAPIES in FRANCE, KARREN in Germany and SWALLOW HOLES in Yugoslavia. Further steepening of the joints have produced solution valleys and caverns. The holy Vaishno Devi Cave is an excellent example of such dilated joints. On cliff faces the joints are numerous and of open type. On the broad rounded ridges, joints are few and tightly closed. On some summits near Hansal (Lat. 32° 59′ 50″ N: Long. 74° 56′ 30″ E) small and sharp protuberances are traversed by irregu-lar intricate and highly curved joints. They are mostly conformable with the trends of the protuberances themselves. Riasi (Lat. 33° 05′ N: Long. 74° 50′ E area has a typical domal structure. It is one of the biggest inliers. The creep of big blocks under the force of gravity, have produced joints which are either parallel or perpendicular to the general trend of the creep. The parallel joints are called slope JOINTS while the perpendicular are designated as CONTOUR JOINTS. The SLOPE JOINTS are radial while the CONTOUR JOINTS are annular.

Bidda (Lat. 33° 07′ N: Long. 74° 49′ 30″ E) has a typical terraced topography. It represents mostly a flat plain constituted of alluvium and river gravels. The joints noticed in the area are mutually perpendicular and thse are the result ofrolling of rough debris along the slopes of the adjoining hills.

Dhiangarh (Lat. 33° 08′ 25″ N: Ling. 74° 48′ E) area has a rugged mountainous topography. The limestone is traversed by an important consequent stream, the CHENAB. The joints in this terrain are either parallel to the bedding or dip 50 in a southerly direction. Most of these joints are the consequence of the crustal deformations. The incipient joints in the limestone are exclusively due to local blasting done in connection with the construction of a local dam.

Origin of Joints

The joints studied in the whole terrain have been mostly filled with vein minerals. Such fillings of the silica are very common at Katra and at Dhiangarh. It is thus obvious that the topography has a great bearing on the orientation of the joints. The spacing, frequency and their characters are greatly influenced by the configuration of crustal topogrphy. The detailed account of the joints controlled by the topography and physiography of the area will be published elsewhere.



FIGS. 1 and 2. Development of joints in the limestone in the Jangal Gali area.

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The author is highly indebted to Dr. T. R. Bhatia for constant guidance and encouragment in the work.

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Jammu (J & K), *July* 30, 1975.

1. Chadha, S. K., *Curr Sci.*, 1975, **44** (18), 665.
2. Chapman, C. A. and Rioux, R. L., *Amer. Jour. Sci.*, 1958, **258**, 3.
3. —, *Journ. Goel.*, 1958, **66** (5), 552.
4. Raha, P. K. and Sastry, M. V. A., *Him. Goel.*, 1973, **3**, 135.
5. Medlicott, H. B., *Mem. G.S.I.*, 3(2), 1864, PL–206.
6. —, *Recd. G.S.I.*, 1875, **9** (2), 1864, 53.
7. Rao, C. Gundu, *Pub. Cent. Adv. Goel.*, 1970, **7**, 127.
8. Wadia, D. N., *Mem. G.S.I.*, 1928, **51** (2) **B**, 253.

CHROMOSOME ORGANIZATION OF *PERIPLANETA AMERICANA*

CHROMOSOME morphology of *Periplaneta americana* has been the matter of discussion from 1909[1]. Classical paraffin section methods enabled[2,3], to conclude that the chromosomes are telocentric. They used testicular material for their observation. John and Lewis[4-6], used female blood cells and testicular material treated with colchicine and made squashes to study the chromosomes and concluded that there are constrictions in the chromosomes-median and/or subterminal. Their microphotographs clearly indicate their claims that the centromere is interstitial and not terminal. Rajasekarasetty and Ramanamurthy[7] made Feulgen squash preparations of Indian *Periplaneta americana* and concluded that most of the spermatogonial metaphase chromosomes are submetacentric and a few are acrocentric. In an unidentified species of *Periplaneta* Rajasekarasetty and Ramanamurthy[8] reported that the kinetochore was either submedian or subterminal. Theirs were mostly camera lucida drawings and the material used was Feulgen squash preparations of the testis. Thus, the position of the kinetochore in the chromosome architecture has not been unequivocally spelled out. Therefore the authors reinvestigated the chromosome morphology of Indian *Periplaneta americana,* using the hepatic caecae of both males and females after colchicine treatment, centrifugation, flame drying the cell suspension and staining with Giemsa. The karyotype was constructed (Fig. 1). The morphometric measurements were made according to Levan *et al.*[9], with minor modifications for all the chromosomes. The idiogram was constructed (Fig. 2) and the L^R and I^c values were calculated and as given in Table II.

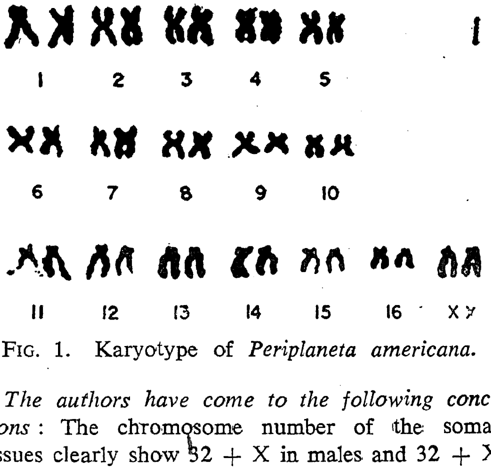

FIG. 1. Karyotype of *Periplaneta americana.*

The authors have come to the following conclusions : The chromosome number of the somatic tissues clearly show 32 + X in males and 32 + XX in females. Thus establishing XX and XO mechanism of sex determination in the cockroach. There is no clearcut distinction between chromosomes with regard to length as they are graded in length (Fig. 2). Even though the microphotographs of John and Lewis[6] are the best that the authors have come across, they did not construct the karyotype nor made the morphometric measurements as their interests were different. Karyotyping of chromosomes of hepatic caecae in the material under study are clearer with respect to the positions of kinetochore.

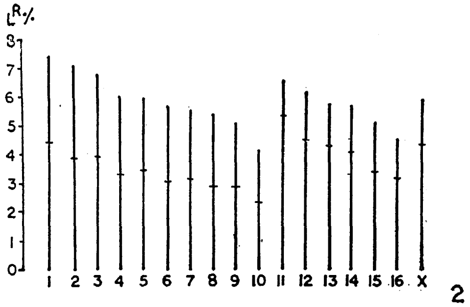

FIG. 2. Idiogram of *Periplaneta americana.*

TABLE I

Classification of metaphase chromosomes after Levan et al. (1964)

Nomenclature	Location	Centromeric index (I^c)
m	median region	37·5–47·5
sm	submedian region	25·0–37·5
st	subterminal region	12·5–25·0
t	terminal region	2·5–12·5

TABLE II

Showing the measurements of chromosome complement of Periplaneta americana

Chromosomes No.	L^R	I^c
1	74·16	39·56
2	71·71	45·45
3	68·45	40·76
4	61·93	44·73
5	60·30	41·89
6	57·90	45·71
7	56·23	43·48
8	54·60	46·26
9	51·35	42·85
10	42·96	41·50
11	66·01	27·16
12	62·75	27·27
13	58·62	25·00
14	56·99	28·27
15	51·28	33·33
16	45·65	30·35
X	59·45	26·02

The nomenclature and morphometric measurements have been made according to the method proposed by Levan *et al.*[9] (Table I). The average of the measurements from 5 plates has shown that the I^c values of the 10 pairs of metacentric chromosomes range from 39·56 to 46·26 and those of the 6 pairs of submetacentric chromosomes and the X-chromosome range from 25·00 to 33·33. So the authors opine that the first 10 pairs are metacentric chromosomes with the kinetochore in the median region (*m*) and the remaining 6 pairs and the X are submetacentric with the kinetochore in the submedian region (*sm*).

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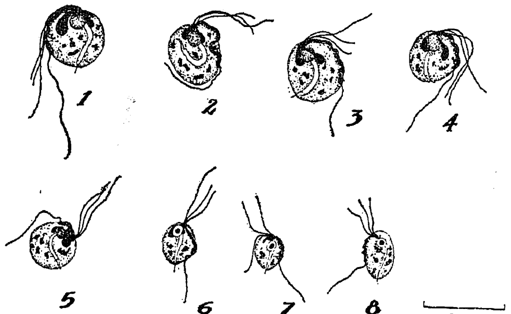
---

1. Morse, Max., *Arch. F. Zelliforsch,* **1909**, **3**, 483.
2. Sharma, G. P., Prasad, R. and Sehgal, P., *Nature,* 178, 1004.
3. —, —. and —, *J. Genet.,* 1959, **56**, 281.
4. John, B. and Lewis, K. R., *Heredity,* 1957, 2, 1.
5. — and —, *Ibid.,* 1958, 12, 185.
6. — and —, *Ibid.,* 1960, 15, 47.
7. Rajasekarasetty, M. R. and Ramanamurthy, C. V., *Nature,* 1963, 197, 1325.
8. — and —, *La Cellule,* 1964, 65, 25.
9. Levan, A., Fredga, K. and Sandberg, A. A., *Hereditas,* 1964, 52, 201.

ON A NEW SPECIES OF THE GENUS *HYPOTRICHOMONAS* LEE, 1960 (SUB-FAMILY : HYPOTRICHOMONADINAE) FROM *HEMIDACTYLUS* SPECIES

FROM the recent studies of morphogenesis, Honigberg[1], 1963, suggested that *Hypotrichomonas* represents a step leading from Monocercomonadidae towards Trichomonadidae because of the structure of its undulating membrane, its axostyle and its parabasal apparatus which are foreshadowed in Monocercomonadidae. He thus anticipated the existence of members of Hypotrichomonadinae with a stout tube-like axostyle, probably equipped with periaxostylar rings and tapering to a point at the postrior end. So far no such flagellate was reported. The present investigation revealed the existence of such flagellate from a lizard *Hemidactylus* sp. from Warangal region, A.P., India.

The new species is almost round in shape (Figs. 1, 3–5). It measures 5·9 μ to 10·9 μ with an average of 8·2 μ in diameter. The blepharoplast, situated at the anterior end gives origin to the four flagella. The three anterior flagella are subequal in length (Figs. 1, 3–4) and measure 5·0 μ–15·4 μ, 5·0 μ–12·9 μ, 5·0 μ–12·3 μ with an average of 10·0 μ, 9·6 μ, 9·1 μ respectively. The posterior flagellum runs backward, attached to the body surface for a considerable length (Figs. 1–8). The attached portion is thrown into a shallow undulating membrane (Figs. 1–5) and the extent of it is variable in different forms. The trailing flagellum measures 5·0 μ–18·1 μ with an average of 12·0 μ in length. The spherical nucleus is situated at the anterior end of the body (Figs. 1–2). It measures 1·0 μ–3·1 μ with an average of 2·0 μ in diameter, and contains a conspicuous central endosome (Figs. 6–8). The axostyle is tubular and always curved in the cytoplasm (Figs. 1, 3–5). The anterior part is obscured by the nucleus and the posterior part is equipped with distinct periaxostylar granules (Figs. 1–4) and abruptly tapers to a point forming a conical projection (Figs. 1–3). Pelta is inverted comma shaped (Figs. 2–4) and parabasal body is bilobed extending on either side of the nucleus (Figs. 1, 3, 4, 6–7).



FIGS. 1–8. *Hypotrichomonas hemidactyli* n. sp. Figs. 1–5. From material fixed in methanol and stained with Giemsa's. Figs. 6–8. From material fixed in Schaudinn's and stained with Heidenhain's iron haematoxylin. All the drawings are made at a magnification of, × 2,200.

Fig. 1. Showing general structure; Figs. 2–5. Showing curved axostyle with periaxostylar ring; Figs. 3–4. Showing subequal flagella, bilobed parabasal body; Figs. 6–8. Showing round nucleus with central endosome.

As far as can be assertained, two species from reptiles and one from bird were described. *H. acosta* (Moskowitz[2], 1951) Lee[3], 1960, is ovoidal in shape, measuring 4·0 μ–17·0 μ × 2·5 μ–15·0 μ. The trailing flagellum is acronematic and the robustus axostyle is distinguishable into spatula and a twished trunk. While the present species is round and differs in size, the trailing flagellum is without acroneme and the axostyle is tubular with a small spike and periaxostylar ring. *H. osmaniae* described by Krishnamurthy[4] (1967) from varanid lizard, is fusiform or pear shaped, measuring 7·2 μ–13·88 μ × 4·11 μ–12·85 μ with an average of 9·94 μ × 6·65 μ. The axostyle is tubular with long spine like projection. The nucleus is elliptical in shape with granular mass. Whereas the present species is round and differs in size, has a tubular axostyle with short spike always curved in the cytoplasm, nucleus is round and has a distinct endosome and the axostyle with periaxostylar granules. An incomplete and inadequate description is given by Geiman and Wichterman[5] (1937) from land tortoises. That organism possesses equal anterior flagella, while the present form has subequal flagella. *H. avium* E. S. Navarathnam (1970), from bird *Cryptoplectron erythrorhynchum*, differs from the present species in having a spherical or pear shaped body, measuring (4·5 μ–9·5 μ) 6·4 μ × (3·5 μ–7·5 μ) 5·5 μ and cytostome.

The presence of periaxostylar ring itself distinguishes it from all hitherto known species of *Hypotrichomonas* and gives it a specific status. Therefore, it is proposed to designate as *Hypotrichomonas hemidactyli* n. sp., after the generic name of the host.

The authors are grateful to Prof. P. Ramchander Rao, for encouragement and laboratory facilities.

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---

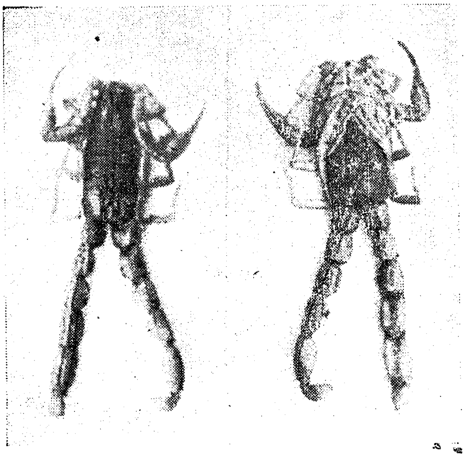
1. Honigberg, B. M., *J. Protozool.*, 1963, 10, 20.
2. Moskowitz, N., *J. Morphol.*, 1951, 89, 257.
3. Lee, J. J., *J. Protozool.*, 1960, 7, 393.
4. Krishnamurthy, R., *Curr. Sci.*, 1967, 21, 583.
5. Geiman, Q. M. and Wichterman, R., *J. Parasitol.*, 1937, 23, 331.
6. Navarathnam, S. E., *Riv. Parassit.*, 1970, 31, 251.

A REPORT ON THE OCCURRENCE OF TWIN-TAILED SCORPION

EXAMPLES of abnormal development of different structures have been described by several investigators[1-4]. However, no case of anomaly in the structures of scorpion is available in the published record. In the present report an unusual occurrence of twin tails in common scorpion has been described.

The specimen was brought to the Department of Zoology from nearby locality of Bhopal. Two complete tails, identical in structure are attached to the base of mesosoma (Figs. 1 and 2). There could not be found any significant difference between the two tails except the tail of the left side is slightly thicker than that of the right side. The segments of the right tail are dorsally concave and ventrally convex with longitudinal ridges and its ventral side is somewhat concave. Correspondingly the right sting is pointed upwards and left downwards.

On careful examination of the specimen there could not be found any other abnormality, except the twin tails. The morphological details of the specimen remain the same as that of a mature normal scorpion.



FIGS. 1 and 2.

As the embryology of this specimen is not studied, no definite explanation for the development of the twin tails could be putforth. However, it is assumed that the cell which initiates the tail formation was split into two equal parts and each part then grew into a fully formed tail.

We are thankful to Prof. M. A. Qayyum, for his constant encouragement and suggestions.

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---

1. Chakraborty, S. and Ghosal, D. K., *Curr. Sci.*, 1971, 4, 88.
2. Hora, S. L., *Rec. Indian Mus.*, 1921, 22, 27.
3. Rajulu, G. S., *Curr. Sci.*, 1970, 24, 564.
4. Tandon, K. K. and Sharma, K., *Ibid.*, 1971, 16, 438.

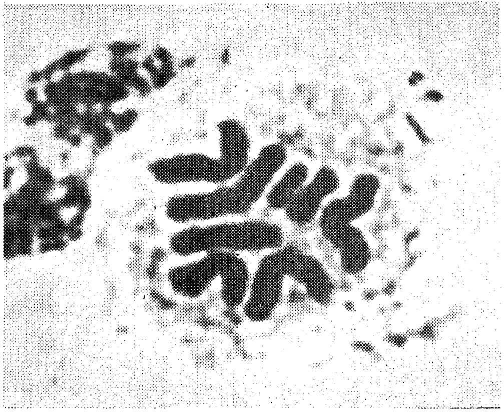
KARYOTYPE OF THE SANDFLY *PHLEBOTOMUS PAPATASI*

SANDFLIES form an important group of vectors of many human diseases. Over 500 species of sandflies of the family Phlebotomidae are known from various parts of the world. Though there are many reports of the studies on ecology, general biology and vector potentiality of sandflies, very little is known about their cytology. In fact, the only published report on this subject is that of the karyotype of *Phlebotomus longipalpis* (*Lutzomyia longipalpis*)[1]. The present communication reports the karyotype of another species of sandfly *Phlebotomus papatasi.*

Fully grown 4th instar larvae from the laboratory colony of *P. papatasi* were used for chromosome preparations. They were immersed in 0·1% colchicine solution for about 4 hours. Following colchicine treatment, the head of the larva was separated from the rest of the body and stained for 10 minutes in lacto-aceto-orecin stain (85% lactic acid, 50 ml; acetic acid glacial, 50 ml; orecin, 2 gm). The whole head after staining, was squashed in 45% acetic acid on a slide. The brain tissue could thus be easily seen. The edges of the cover-slip were sealed with nail-polish and the preparations were studied under microscope. A total of 35 larvae were processed, 23 individually and the rest in batches of 2–3. Metaphase plates were obtained from 6 out of 23 larvae processed individually, in addition to those observed from larvae processed in batches.

The karyotype of *P. papatasi* consists of 4 pairs of chromosomes ($2n = 8$) (Figs. 1–2). The chromosomes are numbered from I to IV according to the system followed in case of mosquitoes[2]. The shortest chromosome is designated as chromosome I, the longest as IV and the intermediate ones as II and III. Of these, chromosome I is acrocentric, chromosomes II and III submetacentric, and chromosome IV metacentric. The relative length of different chromosomes and their arms, from one of the metaphase plates, are as follows: chromosome I—2·8 μm, chromosome II—5·6 μm ($3·6 + 2·0$ μm), chromosome III—7·0 μm ($4·5 + 2·5$ μm) and chromosome IV—7·6 μm ($3·8 + 3·8$ μm). Sexual dimorphism in karyotype could not be detected in any of the metaphase plates studied.

Carvalho[1], reported for the first time, the karyotype of the sandfly *P. longipalpis,* which also had 4 pairs of chromosomes. Of these, one was acrocentric (chromosome I) and the remaining three were metacentric (chromosomes II, III and IV) pairs. Whereas, in *P. papatasi,* as it has been observed during the present study, there is one acrocentric pair (chromosome I) and one metacentric pair (chromosome IV) and the other two submetacentric pairs (chromosomes II and III). It is thus evident that the karyotypes of these two species of sandflies, one from the new world (*P. longipalpis*) and the other from the old world (*P. papatasi*) differ considerably.



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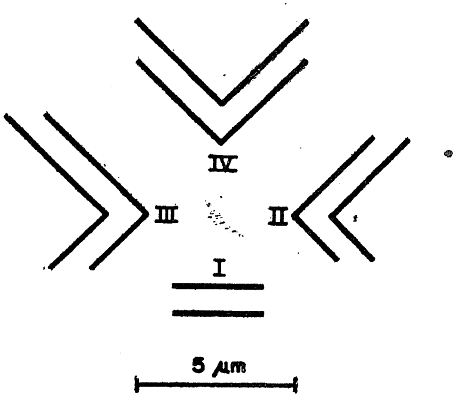

2

FIGS. 1–2. Fig. 1. Metaphase chromosomes of *Phlebotomus papatasi*, × 1,500. Fig. 2. Diagrammatic representation of the karyotype of *Phlebotomus papatasi*.

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---

1. Carvalho, H. C., Falco, A. and Schrelber, G., *Ciencia e Cultura,* 1962, 14, 38.
2. Rai, K. S., *Ann. Entomol. Soc. Amer.,* 1963, 56, 160.

KARYOTYPES OF THREE SPECIES OF MOSQUITOES FROM INDIA

CYTOGENETICS of mosquitoes has received much attention in recent years in an attempt to understand the genetic basis of insecticide resistance and genetic control. Even though more than 2400 species of mosquitoes are known to science so far, various cytogenetic studies have been conducted on less than 100[1-4]. Kitzmiller[2] emphasized the need for standardized karyotype studies in additional species of mosquitoes to obtain data on heterosomes and relative arm length of mitotic chromosomes. The purpose of this communication is to report the karyotypes of three species of mosquitoes from India, *viz.*, *Aedes* (*Stegomyia*) *novalbopictus* Barraud, *Aedes* (*Stegomyia*) *subalbopictus* Barraud and *Toxorhynchites splendens* (Wiedemann).

Brain tissues from the 4th instar larvae were employed to study the somatic chromosomes. Larvae of *A. novalbopictus* and *A. subalbopictus* were from approximately 35 and 5 laboratory generations respectively, whereas larvae of *T. splendens* were collected from the field (Khandala, Maharashtra State) and were reared in the laboratory up to 4th instar. The techniques employed for chromosome preparations were essentially the same as described by French *et al.*[5]. From each species 25–30 individual larval brain preparations were studied. For chromosome measurements the procedure used by Rai[6], was followed. They were numbered according to McDonald and Rai[7].

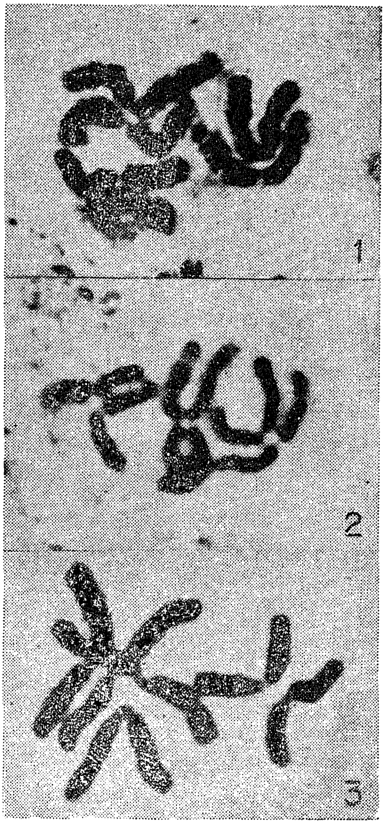
The diploid chromosome number was 6 in all 3 species (Figs. 1–3). Typical of mosquitoes, chromosome were arranged in 3 pairs. Occasionally (0·1–0·2% of the dividing cells) cells with polyploid chromosomes were also observed in all the 3 species. Somatic pairing of homologous chromosomes was frequent and was intimate at the centromere region. Sexual dimorphism in the chromosome complement was not detected in any of the species studied. The measurements of metaphase chromosomes and the ratio of the length of chromosome I to chromosomes II and III are given in Table.

All the chromosomes from 3 species of mosquitoes were metacentric. The ratio of length of chromosome I to chromosomes II and III in *A. novalbopictus* and *A. subalbopictus* was comparable to that of the other members of the genus *Aedes* thus far studied[6,8,9]. The ratio obtained for *T. splendens* was closer to that of members of the genus *Aedes* than to members of the genera *Culex* and *Anopheles*. Both in *A. novalbopictus* and *A. subalbopictus* there was a considerable difference in length between chromosomes II and III (6–7% of the total length

TABLE
Measurements of metaphase chromosomes in 3 species of mosquitoes from India

Species	Chromosome I* (small)			Chromosome II* (large)			Chromosome III* (medium)			Ratio of length of I to II + III
	p.c.	arm a	arm b	p.c.	arm a	arm b	p.c.	arm a	arm b	
Aedes novalbopictus	M	5·5	5·5	M	7·7	7·7	M	6·7	6·7	0·382
Aedes subalbopictus	M	3·3	3·3	M	4·8	4·8	M	4·2	4·2	0·367
Toxorhynchites splendens	M	4·8	4·8	M	7·0	7·0	M	6·7	6·7	0·350

* all measurements in μ m. p.c.—position of centromere. M—metacentric.



FIGS. 1–3. Mitotic chromosomes from brain cells, lacto-aceto-orecin stained. Fig. 1. *Aedes novalbopiltus*, × 2,500. Fig. 2. *Aedes subalbopictus*, × 4,000. Fig. 3. *Texorhynchites splendens*, × 3,000.

of 6 chromosomes), whereas in *T. splendens* this difference was much less (2% of the total length of 6 chromosomes). The variation thus observed in the comparative length between the chromosomes in mosquitoes belonging to different genera might be of evolutionary and genetic significance. However, further detailed comparative studies on the karyotypes of additional species of mosquitoes are indicated.

I wish to thank Dr. N. P. Gupta, Director, and Dr. V. Dhanda, Assistant Director, for their encouragement during the course of the present study. My sincere thanks are also due to Mr. S. N. Guttikar of the Entomology Division, for his assistance in rearing *A. novalbopictus* and *A. subalbopictus* and for field collection and identification of *T. splendens* larvae.

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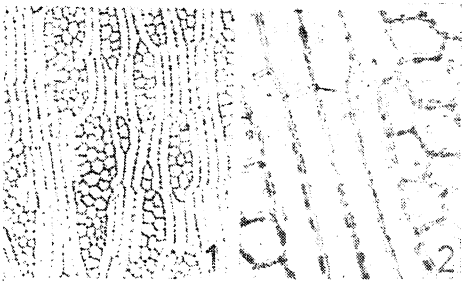
1. Kitzmiller, J. B., *Bull. Wld. Hlth. Org.*, 1963, 29, 345.
2. —, *Genetics of Insect Vectors of Disease*, Ed., Wright, J. W. and Paul, R., Elsevier Publ. Co., Amsterdam, 1967, p. 133.
3. Davidson, D. and Mason, G. F., *Ann. Rev. Ent.*, 1963, 8, 177.
4. WHO. Tech. Rep. No. 268, 1964.
5. French, W. L., Baker, R. H. and Kitzmiller, J. B., *Mosquito News*, 1962, 22, 377.
6. Rai, K. S., *Ann. Ent. Soc. Am.*, 1963, 56, 160.
7. McDonald, P. T. and Rai, K. S., *Genetics*, 1970, 66, 475.
8. Mukherjee, A. B., Rees, D. M. and Vickery, R. K., *Mosquito News*, 1966, 26, 150.
9. Singh, K. R. P. and Bhat, U. K. M., *Curr. Sci.*, 1970, 39, 537.

ON THE OCCURRENCE OF STRATIFIED CAMBIUM IN *SALVADORA PERSICA* L.

FOLLOWING Bailey[1], two types of cambia are recognized in the various plants showing secondary growth, namely storied or stratified and non-storied or non-stratified. The former is recognized by the uniform length of the fusiform initials whereas in the latter these acquire variable dimensions. Phylogenetically, the latter is considered as primitive, since it occurs in the fossil representives of the lower groups including the gymnosperms. The storied cambium is, however, recorded only in comparatively fewer taxa of the angiosperms. The purpose of this note is to record the occurrence and organization of the storied cambium, an additional interesting feature for *Salvadora persica* L.



FIGS. 1–2. Fig. 1. Cambium of *S. persica* in tangential section showing storied arrangement of fusiform initials, × 130. Fig. 2. Enlarged portion of Fig. 1. showing the beaded appearance of the fusiform initial cell walls and the nuclei in the ray and fusiform initials, × 550.

During the survey of the cambial organization and its seasonal activity in a number of tropical trees a common indigenous tree *Dalbergia sissoo* was reported to possess a storied type of cambium by Paliwal and Prasad[4]. This was later confirmed by Ghouse and Yunus[3], as a substitute material for *Robinia pseudoacacia,* the well-known European tree exhibiting this feature. More recently the same authors have listed more plants, common to northern India possessing the storied cambium.

Our material has been collected from a tree growing at the Old Delhi Ridge, from the main tree trunk at breast height. Small pieces of wood (1½ × 3 cm), containing bark were fixed in craf III for 24 hrs. They were finally preserved in 70% alcohol after washing thoroughly with water. Transverse and longitudinal sections were cut at 15–20 µm with the help of Reichert Sliding Microtome. The sections were passed through tannic acid-ferric chloride-lacmoid series as outlined by Cheadle *et al*[2]. Mounting was done in Eukitt.

As seen in tangential longitudinal sections, the fusiform initials are shorter and arranged in regular tiers one above the other. The cell walls of the fusiform initials showed beaded appearance due to the primary pit fields. These are uninucleate and highly vacuolate. Divisions occur in them by means of radical longitudinal walls. The rays are uni- to multiseriate and isodiametric in outline. During the span of a year, the length of the fusiform initials varies from 120 µm to 156 µm and the breadth from 12 µm to 16 µm. The dimensions of the ray initials range from 14·5 µm to 20·0 µm.

We express our gratitude to Professor H. Y. Mohan Ram, the Head, Department of Botany, for encouragement and laboratory facilities and the UGC, for the award of a Junior Research Fellowship at the Centre of Advanced Study in Botany of the University of Delhi to one of us (V. S. S.).

Plant Anatomy Laboratory, V. S. SAJWAN.
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Delhi 110 007, India, *August* 19, 1975.

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1. Bailey, I. W., *Am. J. Bot.,* 1923, 10, 499.
2. Cheadle, V. I., Gifford, E. M. Jr. and Esau, Katherine, *Stain Technol.,* 1953, 28, 49.
3. Ghouse, A. K. M. and Mohd. Yunus, *Curr., Sci.,* 41, 569.
4. Paliwal, G. S. and Prasad, N. V. S. R. K., *Ibid.,* 1971, 40, 114.

STALK END ROT OF GRAPES

DURING months of April and May 1975 a stalk end rot of grapes was observed at Allahabad. Percentage infection of the fruit varied from 3 to 5.

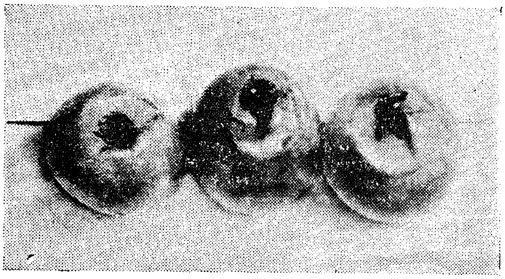

FIG. 1. Stalk-end rot of grapes.

Affected berries showed brownish flaccid rot at the stalk-end. The affected fruits fall off with the slightest touch to the bunch. The rot starts as a small circular to oval water-soaked spot which enlarges and becomes brown with time. White mycelia of the fungus develop on the centre of the diseased area and later black conidial heads of the fungus develop. Affected tissues become pulpy and shrink. The rotted fruit emit a fermented odour.

Isolations made from the diseased tissues as well as from the mouldy growth revealed the presence of *Aspergillus niger* van Tieghem. Pathogenicity tests were made by spraying the conidial suspensions on pricked berries which developed rot. The fungus closely resembles with description of *A. niger* as described by Raper and Fennell[1]. *A. niger* is well known to be present in the atmosphere, storage places and also on the surface of various fruits[2] and therefore it is advisable that grapes should be picked and preserved avoiding injuries to the berries.

Rot of grapes due to *A. carbonarius* (Bain.) Thom has been already reported[3], but this is a new record of *Aspergillus niger* on the berries of *Vitis vinifera* L. from India.

Authors are grateful to Prof. D. D. Pant, Head of the Botany Department, University of Allahabad, Allahabad, for providing necessary laboratory facilities.

Botany Department,
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A. P. Singh.

1. Raper, K. B. and Fennell, D. I., *The Genus Aspergillus*, The Williams and Wilkins, Co., Baltimore, 1965, p. 686.
2. Sinha, S., *Proc. Ind. Acad. Sci.*, 1946, 24 B, 198.
3. Gupta, S. L., *Sci. and Cult.*, 1956, 22, 167.

ABNORMAL FEMALE CONES OF *ZAMIA FLORIDANA* A. DC. (CYCADACEAE)

Zamia floridana A. DC., a native of Florida, cultivated in the Experimental Garden of the Botanical Survey of India, Central Circle, Allahabad, produced both normal and abnormal female cones on a single plant. The plant under study produced eight cones. Out of these, six were normal and two abnormal in external morphological characters.

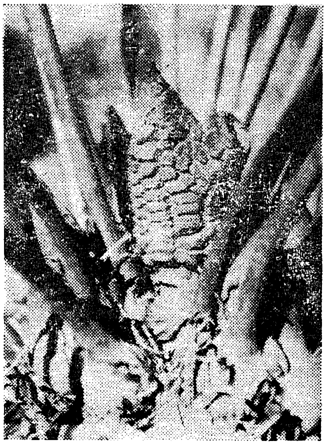

Fig. 1. *Zamia floridana* A. DC. shows bifurcated cone.

Occurrence of multiple cones with normal constituents/individual cones on a single plant of *Zamia floridana* A. DC., have already been reported by Smith (1929), Coulter and Chamberlain (1964) and Graph (1973).

One of the abnormal cones present at the crown exhibited normal middle and distal portions, whereas proximal end showed bifurcation (Fig. 1). The bifurcation originated 1·5 cm below the apex of the cone and directed upward. Megasporophylls were 0·4 cm × 0·2 cm in size and hexagonal in shape.

The second abnormal cone was also normal in the middle and distal end like the first one but trifurcated at the proximal end. Megasporophylly were 0·8 cm × 0·3 cm in size and hexagonal in shape. Ovules were of normal type in both these cones. The growth of the trunk was noticed to be sympodial and the vascular supply continued in the furcations from the main axis.

The normal and abnormtl cones, preserved in F. A. A., are kept in the museum of the Botanical Survey of India, Central Circle, Allahabad.

The authors are grateful to Dr. B. D. Sharma, Regional Botanist, Botanical Survey of India, Central Circle, Allahabad, for kindly going through the manuscript.

Botanical Survey of India,
Central Circle,
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1. Coulter, J. M. and Chamberlain, C. J., *Morphology of Gymnosperms*, Allahabad, 1964.
2. Graph, A. B., *Exotica : Pictorial Cyclopedia of Exotic Plants*, Rutherford, N. J. (U.S.A.), 1973.
3. Smith, F. G., *Bot. Gazette*, 1929, 88, 204.

HARKNESSIA : A NEW GENERIC RECORD TO INDIA

While collecting fungi at Khandala, the author encountered an interesting pycnidial fungus on leaves of *Memecylon edule* Roxb., which was identified as a species of the genus *Harknessia* Cooke apud Cooke and Harkness. The genus was established by Cooke with *H. eucalypti* Cooke as type species in 1881[1]. Sutton[2], while studying this genus accepted sixteen species. Critical comparative study revealed that, the present species is new to science and is described in this communication.

Harknessia gharsii Golatkar Sp. nov. (Fig. 1). Fructificationes abundantes, discretae, epiphyllae, immersae, subepidermates, globosae, uniloculares, erumpentes, usque ad 200 μ diam. Parties basales et laterales stromatum 3–4 cellulis crassis, excellulis subhyalinis vel pallide brunneis psudoparenchymaticis compositi, qui sunt hyalinis versus partorn cellulorum conidigenorum; ostioli 33 μ distncti et margines furfurace absentes, aperturas stromatibus lata et plus minusve circularis, cellulae conidiogenae ex cellulis intericribus parietis stromatibus formatae, longae langeniformae, ad bases 5 μ latae, et interdum 0-septatae, hyalinae, laves, nonramosae 40 μ longae. Conidia (blastosporae) aseptata, midiobrunnae, laevia, 13·33–16·66 × 6·66–10·0 μ, cum appendice hyaline apiculi, 1·0–1·5 μ longa, basis bruncata cum appendice minuta.

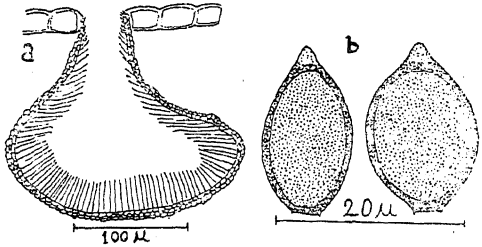

FIG. 1. *Harknessia gharsii* Golatkar. *a*, Pycnidium in section; *b*, Conidia.

In follis viventibus *Memecylon edule* Roxb., Khandala (Maharashtra State), 15/8/1974, V. V. Golatkar, typus: A M H 2629.

The present species differs from all the accepted species except *H. insueta* Sutton, in having basal appandage reduced to a marginal frill. In *H. insueta* Sutton the conidia are oblique and napiform, while in the present species they are symmetrical and elliptical. These differences justify its inclusion in a new proposed species.

This is the first report of the genus *Harknessia* Cooke apud Cooke and Harkness from India. Specific epithet commemorates Dr. P. S. Gharse, Head, Biology Department.

The author is grateful to Dr. P. S. Gharse, for his valuable guidance and constant encouragement.

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1. Cooke, W. M., *Grevillea*, 1881, 9, 85.
2. Sutton, B. C., *Mycol. Pap.*, 1971, 123, 1.

PROLONGED PRESENCE OF NUCLEUS IN THE SIEVE TUBE ELEMENTS OF CHILLI AND BRINJAL

SOME interesting observations on nucleus in sieve tube elements in the stem of chilli and brinjal, two solanaceous species, were recorded during the detailed anatomical studies[1]. The dispersal of slime or P-protein bodies and the disorganization of the nucleus occur more or less concomitantly in many angiosperms[2-5]. However, in the brinjal and the chilli, the nucleus remains in a sieve element even after the dispersal of P-protein body. In *Neptunia oleracea* and *Nelumbo nucefera* the nucleus is reported to be present in a seive element for a brief time even after the formation of the sieve plate[6,7]. The nucleus is retained in a sieve element even after the formation of seive plate and formation of slime or P-protein plug in chilli and brinjal. The nucleus in the differentiating sieve element is spherical and chromophilic. The normal appearing nucleus is present during the formation of the sieve plate (Fig. 1 A). Later, the nucleus elongates (Fig. 1 B). Its presence during the P-protein body formation (Fig. 1 B) and dispersal, and plug formation (Fig. 1 C) is of particular interest. The nucleate sieve elements are found in an elongating internodes. Their retention in sieve tube elements for a long time suggests that the nucleate elements could be functional, and enucleate ones must be functional. Multinucleate sieve elements, as reported in tobacco[8], are not found in chilli and brinjal.

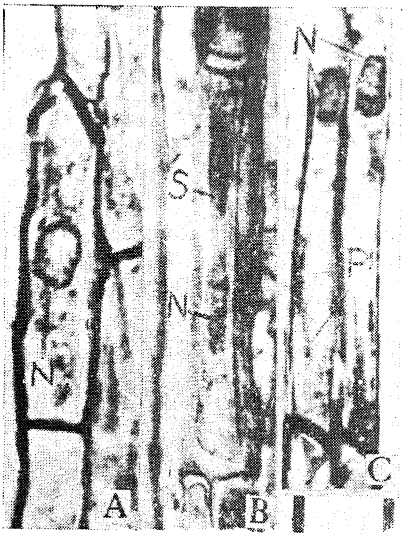

FIG. 1. A–C. Longisections of sieve tube elements. A, B, *Solanum melongena*. C, *Capsicum annuum*. Scale 10 microns. *N*, nucleus; *S*, P-protein body; *P*, slime plug.

My sincere thanks are due to Professor J. J. Shah for his guidance and useful suggestions.

37, Krishna Society, J. D. PATEL.
Anand 388 001, *August* 4, 1975.

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1. Patel, J. D., *Ph.D. Thesis,* S. P. University, Gujarat, 1970,
2. Esau, K., *Bot. Rev.,* 1939, 5, 373.
3. Shah, J. J. and Jacob, R., *Amer. J. Bot.,* 1969, 56, 821.
4. — and —, *Ann. Bot.,* 1969, 33, 855.
5. Esau, K., *The Phloem, Encyclopedia of Plant Anatomy,* Gebruder, Berlin, 1969.
6. Shah, J. J. and James, M. R., *Aust. J. Bot.,* 1968, 16, 433.
7. — and —, *Ann. Bot.,* 1969, 33, 185.
8. Craft, A. S., *Bot. Gaz.,* 1934, 95, 592.

THE MINIMAL TIME REQUIRED FOR NEMATODE EXTRACTION BY OOSTENBRINK'S ELUTRIATOR

A SERIES of extraction trials was carried out for the root-knot nematode, *Meloidogyne incognita* (Kofoid and White, 1919), Chitwood, 1949, from tea soils using the standard Oostenbrink's Elutriator[1], except that in the last stage of sieving the cotton wool filter was replaced by a nylon sieve with a pore size of 35 μ. In each of the 11 replicated extraction experiments, 100 g of soil was processed. The nematodes recovered through the nylon sieves were counted on the first, second, third and sixth hour, and thereafter on 18th, 21st and 24th hour in different sets. Only in one experiment a few nematodes were noticed at the end of 24th hour. Nematodes other than *M. incognita* were also found, but this note concerns only with the larvae of *M. incognita.*

Results presented in Fig. 1 show that in all cases within the first three hours about 86% of the total extractable population came out. Thereafter the extractable populations were very small. A statistical analysis of the data by Ratio test[2], indicated only a 4% variability in the overall extraction efficiency in the first three hours.

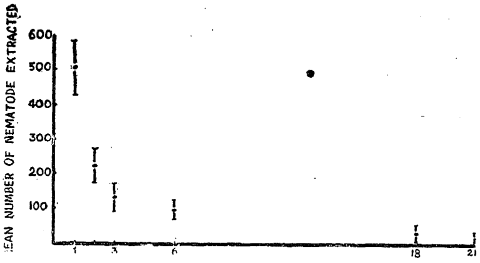


FIG. 1. Average Numbers of *Meloidogyne incognita* Chitwood extracted at different hours by Floatation technique. (The vertical lines are the standard errors of the mean).

To confirm these findings known populations of the second instar larvae of *M. incognita* in water suspension were also processed through the nylon sieves. Results presented in Table I are in agreement with what was noted earlier. In this case the extraction was in fact better than processing of soil samples, as in all cases, the extractable population came out within two hours.

TABLE I

Hourly extraction of introduced population of Meloidogyne incognita *(Kofoid and White, 1919) Chitwood, 1949*

Replication (1500 larvae in each replicate)

Replications / Hours	1	2	3	4	5
1	640	1160	1280	1100	1020
2	60	40	100	80	120
Total	700	1200	1380	1180	1140

No nematode extraction at 3, 4, 18, 21 and 23 hr intervals.

We thank the Director, Tocklai, Experimental Station, for his interest and Tea Research Association, for permission to publish this report.

Tocklai Experimental Station, B. BANERJEE.
Jorhat 8, Assam, S. D. BASU.
July 3, 1974.

---

1. Oostenbrink, M., *Nematology,* Ed., by J. N. Sasser and W. R. Jenkins, Univ. N.C. Press, Chapel Hill, 1960, p. 85.
2. Cochran, W. G., *Sampling Techniques,* John Wiley and Sons, New York, 1953, p. 125.

OCCURRENCE OF THERMOPHILIC FUNGI IN COAL-MINE SOILS OF MADHYA PRADESH

THERMOPHILIC AND THERMOTOLERANT molds occur in a variety of habitats[1,3,4,11,12]. Presence of these organisms under Indian conditions has, however, not been explored inspite of their economic potential as producers of amylases and cellulases[2,5,6,8,12]. During the course of an extensive study of coal-mine soils of Madhya Pradesh several new additions to Indian fungal flora were noted and these alongwith the other notable features of this investigation are reported herein.

Soils collected from the colliery area of District Chhindwara, in M.P. were plated on a variety of media using the plate method[13]. In order to facilitate the appearance of thermophilic fungi, soils were amended with glucose, cellulose, asparagine, ammonium nitrate, or ammonium phosphate and incubated at 45° C prior to plating. Alternatively, unamended soil was incubated at 45° C and samples were plated on ten enrichment media[1,12]. The cultures were maintained on peptone-dextrose and Emerson's YpSs agar media[12]. The temperature relationships were studied by incubating the fungal culture in the range 25–60° C. The identity was confirmed by referring the cultures to CMI, Kew; their depository numbers are cited in the parentheses in the text.

A total of fourteen fungi were isolated.

PHYCOMYCETES: *Absidia corymbifera* (Cohn) Saccardo *et* Trotter (188054); *Rhizopus microsporus* van Tieghem (188056); *Rhizopus rhizopodiformis* (Cohn) Zopf (188055).

ASCOMYCETES *Achaetomium macrosporum* Rai, Wadhwani *et* Tiwari (188068); *Emericella nidulans* (Eidam) Wint (188077); *Thermoascus aurantiacus* Miehe (188061); *Thielavia minor* (Rayss and Borut) Malloch *et* Cain (188066).

DEUTEROMYCETES: *Aspergillus fumigatus* Fresenius (188073, 188078, 188079); *Humicola grisea* Traaen (188076); *Penicillium* sp. I; *Penicillium* sp. II; *Sporotrichum* sp. (188065); *Thermomyces langunosus* Tsiklinsky (188060); *Torula thermophila* Cooneey *et* Emerson (188067).

Amongst these, *A. fumigatus* and *E. nidulans* were found to be thermotolerants, *i.e.,* they could grow at room temperature (22° C) and also at 45° C but with an optimum at 35–40° C. Other molds grew best at 45° C and could also stand a temperature of 55–60° C.

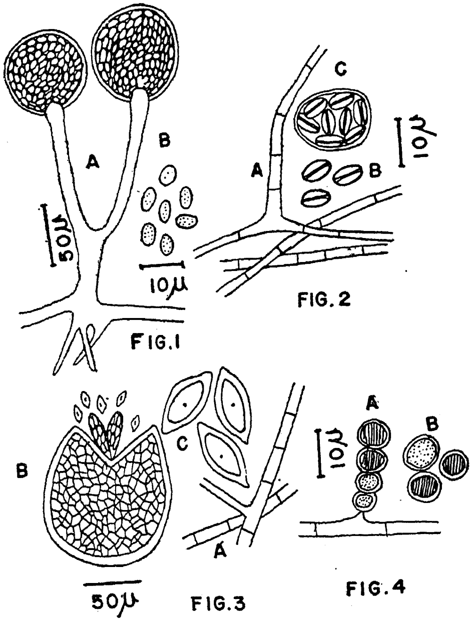
Rhizopus rhizopodiformis, Thermoascus aurantiacus and *Torula thermophila* are new additions to the Indian fungal flora. The species of *Sporotrichum* recovered from coal-mine soils is new. *Achaetomium macrosporum* is known only as a mesophilic fungus but we have isolated a strain which shows strong thermophily and further studies will be required to define its proper taxonomic grouping.

Rhizopus rhizopodiformis (Fig. 1).—Grows very rapidly at 45° C on PDA filling a 9 cm Petri dish in nearly 24 hr. The wooly turf at first white, later turning greyish-black. Vegetative hyphae subhyaline, 3–7 μ diam; stolons somewhat reduced. Sporangiophores single, in twos or in groups from reduced nodal regions; rhizoids pale brown, 122–150 × 12–14 μ. Sporangia, black, spherical, smooth, 76–175 μ diam. Sporangiospores, pale brown, smooth, spherical, 3–6 μ diam.

Thermoascus aurantiacus (Fig. 2).—This fungus grows well on Sabouruad's dextrose agar and PDA, filling a 9 cm Petri dish in 2–3 days at 45° C. The colony appears creamy-white; mycelium of septate hyphae, 2·5–3·5 μ diam; cleistothecia, bright orange to brick red, scattered all over the surface. Asci bright orange, oval to irregular, 11·4–20·0 × 7·6–15·2 μ. Ascospores hyaline, elliptical, 3·8–7·6 × 1·9–5·7 μ.

This organism has been reported from the upper layers of dry pasture soils[2] and from self-heated wood chip piles[12]. It is a new addition to the fungi of India.

Thielavia minor (Fig. 3).—Colony on Emerson's YpSs agar broadly spreading, composed of white, cottony, aerial and submerged hyphae which are 2–5 μ diam. Cleistothecia globose to spherical, walls brown, without ostiole and appendages, 80–160 μ diam, brownish to almost black at maturity. Ascospores, broadly fusiform to elliptical, slightly apiculate at both the ends, dark olivaceous to brown, 10–12·5 × 13–15 μ.



FIGS. 1–4. Fig. 1. *Rhizopus rhizopodiformis.* A, Rhizoidal system, mycelium, sporangiophore and sporangia; B, Sporangiospores. Fig. 2. *Thermoascus aurantiacus.* A, Branched, septate mycelium; B, Asci with ascospores; C, Ascospores. Fig. 3. *Thielavia minor.* A, Branched, septate mycelium; B, Cleistothecium; C, Ascospores. Fig. 4. *Torula thermophila.* A. Mycelium with chains of chlamydospores; B, Thin and thickwalled chlamydospores occurring on the same chain.

Several species of the genus *Thielavia*: *T. sepedonium, T. thermophila,* etc., are thermophilic. Our isolate closely resembles the type described by Evans[3], but differs in that the size of the cleistothecia and ascospores are smaller. A fungus resembling *T. minor* was reported by Lodha[7] and Padmavathy and Rao[9], as *T. terricola* var. *minor* but the coalmine isolate shows strong thermophily and this justifies its placement as *T. minor*.

Torula thermophila (Fig. 4).—Grows well on yeast-malt extract agar, attaining 80 mm diam 3–4 days at 45° C. Mycelium dark brown; hyphae 3·8–6·0 μ diam, septate, cells 3·8–10·0 μ in length. Chlamydospores smooth, dark brown, in long or short chains, usually intercalary, occasionally terminal, 15·2–16·0 × 11·4–13·2 μ. Thin-walled chlamydospores often associated with the chain of dark spores; the former are generally smaller in diam than the latter.

We thank Prof. S. B. Saksena, Head of the Botany Department, for laboratory facilities and Director, C.M.I., Kew, for ideniification of the cultures.

Department of Botany, R. P. THAKRE.
University of Saugar, B. N. JOHRI.
Sagar 470 003, M.P.,
September 8, 1975.

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1. Apinis, A. E., *Nova Hedwigia*, 1963, 5, 57.
2. Eggins, H. O. W. and Malik, K. A., *Antonie von Leeuw. J. Microbiol. Serol.*, 1969, 35, 178.
3. Evans, H. C., *Trans. Br. mycol. Soc.*, 1971, 57, 241.
4. Fergus, C. L., *Mycologia*, 1969, 61, 120.
5. —, *Ibid.*, 1964, 56, 267.
6. Hedger, J. N. and Hudson, H. J., *Trans. Br. mycol. Soc.*, 1974, 62, 129.
7. Lodha, B. C., *Jour. Ind. Bot. Soc.*, 1971, 65, 196.
8. Ofosu-Asiedu. A. and Smith, R. S., *Mycologia*, 1973, 65, 87.
9. Padmavathy, K. and Rama Rao, P., *Indian Phytopath.*, 1970, 23, 589.
10. Sarbhoy, A. K., Girdharilal, and Varshney, J. L., *Fungi of India* (1967–71), Navyug Traders, New Delhi, 1975, p. 148.
11. Taber. R. A. and Pettit, R. E., *Mycologia*, 1975, 67, 157.
12. Tansey, M. R., *Ibid.*, 1971, 63, 537.
13. Warcup, J. H., *Nature, Lond.*, 1950, 166, 117.

VEIN ENATION: A NEW VIRUS DISEASE OF MAIZE IN INDIA

SYMPTOMS of vein swelling and vein enations or galls on the leaves of maize have been observed every year since 1972 in Darjeeling hills. Affected plants were stunted and showed leaf chlorosis. The root system of the diseased plants was much reduced and rotted. Affected plants developed partally sterile tassels which usually did not emerge due to curling or twisting habit of the leaves. The plants developed abnormal cob and the grain yield is reduced. Premature death of many of the affected seedlings was not uncommon. The incidence of the disease ranged from 1 to 15% in different fields. The disease was observed to be spreading in the fields, hence we suspected that the symptom-inducing factor was infectious in nature, possibly a virus, as no pathogenic fungus or bacterium was isolated from the affected plants. Since the disease appeared to cause considerable loss to the maize crop, investigations were undertakes to characterize the causal agent.

The disease was not transmissible by sap, seed or aphids (*Myzus persicae* and *Rhopalosiphum maidis*) but is transmitted by a jassid vector, *Cicadulina mbila* Naude. For transmission studies, insects were collected from the fields and were maintained for a week on diseased and subsequently for four days on healthy seedlings of maize var. Kalimpong local. In the first attmpt 15 out of 20 plants inoculated with *C. mbila* were infected. Transmission of the disease was confirmed repeatedly in experiments using *C. mbila* as vector and glasshouse infected maize plants as virus source.

Symptoms of the disease in glasshouse infected maize plants appeared as white spots on leaf veins. The veins on the lower surface of leaf blade showed severe swelling and numerous white spindle shaped galls or enations usually developed within 3 to 5 days after infection (Fig. 1). Rest of the symptoms were similar to those observed in the field.

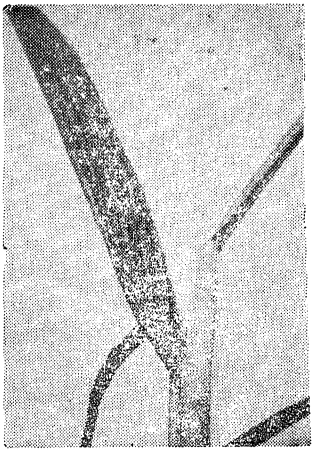

FIG. 1. Maize leaf showing symptoms of vein enation virus.

Various plant species were raised in the glasshouse and inoculated at 4 to 6 leaf stage for host range studies. For testing each species/variety 20 to 30 insects were employed. The insects were given an acquisition feeding of 7 days and a minimum tranmission feeding of 24 hours. Plants that were infected are :

Zea mays (43/50), *Triticum aestivum* (17/40), *Sorghum bicolor* (19/40), *Oryza sativa* (17/28), *Avena sativa* (29/40), *Saccharum officinarum* (4/11), *Eleusine coracana* (20/25), *Setaria glauca* (27/38), *Eleusine indica* (35/49), *Paspalum sanguinale* (17/40), and *Coix lachryma-jobi* (15/27).

The figures in perenthesis represent the number of plants infected out of the number inoculated. Symptoms in all the hosts were similar to those of maize except in sugarcane where only faint swelling of the leaf veins was observed. Fig. 2 shows typical symptoms on wheat.

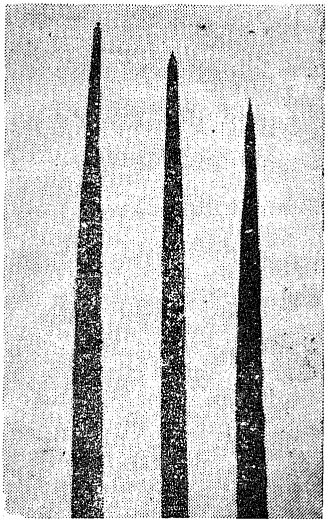

FIG. 2. Wheat leaves affected with MVEV.

In vivus-vector relationship studies, it has been established that both the nymphs and adults were capable of transmitting the disease. The insects could acquire the virus within 15 to 20 minutes feeding on diseased plants. The incubation period of the virus in its vector, *C. mbila* was about 20 hours. Once the insects became viruliferous, they transmitted the disease throughout their lives.

The present virus seems to resemble to maize rough dwarf virus[3] (MRDV), fiji disease of sugarcane[5] (FDV), rice dwarf virus[4] (RDV), maize wallaby-ear disease[6] (MWED), and rice and corn leaf gall virus[1,2] (RCLGV) in symptomatology. RDV, RCLGV and MWEV are transmitted by Cicadellids, whereas MRDV and FDV have delphacid vectors. However, present virus differs from DRV in host range and having a different vector. The enation disease also differs from MWEV, RCLCV in its symptomatology on many new disease with could be designated as vein *C. mbila*. It is, therefore, concluded that this is a common hosts and having a different vector, enation disease.

The authors are thankful to Dr. T. Ishihara, Professor of Entomology, Entomological Laboratory, Ehime University, Matsuyama, Japan, for the identification of the insect vector. We are also grateful to Dr. V. V. Chenulu, Professor of Plant Pathology, IARI, New Delhi, for going through the manuscript and his valuable suggestions.

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1. Agati, J. A. and Calica, C., *Philipp. J. Agric.*, 1949, 14, 31.
2. — and —, *Ibid.*, 1950, 15, 249.
3. Biraghi, A., *Ann. Sper. agr. N.S.*, 1952, 6, 1043.
4. Fukushi, T. J., *Fac. Agric. Hokkaido. Imp. Univ.*, 1934 *a*, p. 37.
5. Lyon, H. L., *Bull. Exp. Sta. Hawaiian Sugar Planter's Assoc. Bot. Ser.*, 1921, 3, 1.
6. Schindler, A. J. J., *Austral, Inst. Agric. Sci.*, 1942, 8, 35.

MORPHOLOGICAL AND HISTOCHEMICAL STUDIES IN NATURAL AND CHEMICALLY INDUCED MALE-STERILE PLANTS

STUDIES on mechanism of male sterility in natural (*ms*) and chemically induced male-sterile (*ims*) plants of *Allium cepa, Beta vulgaris, Capsicum annuum, Cucurbita maxima, Datura alba, Ranunculus scleratus, Sesamum indicum, Solanum melongena* and *Triticum aestivum* were undertaken.

Morphological and cytological studies have revealed that pollen abortion in the above mentioned *ms* and *ims* plants is associated with the following types of abnormal behaviour of tapetum :

1. Tapetal cells degenerate or become hypertrophied in pre-meiotic stages in the central flower of the spikelets of *ms Triticum aestivum* having *Aegilops ovata* and *caudata* cytoplasm and *ms Cucurbita maxima*. In the former, the tapetal cells start degenerating much before the onset of meiosis in pollen mother cells. This is followed by the complete degeneration of both the tissues. In *ms Cucurbita maxima*, the tapetal cells remain intact and their cells become hypertrophied and crush the

pollen mother cells much prior to their meiotic division.

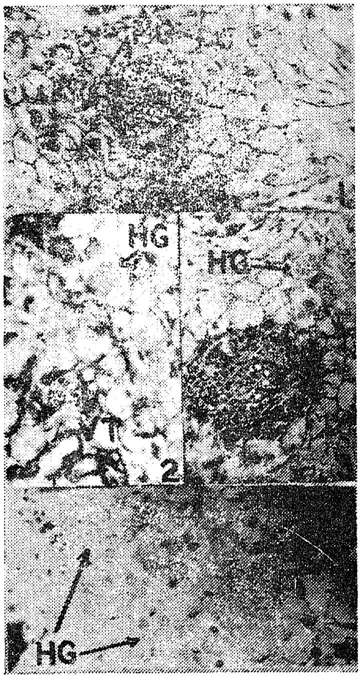
2. Tapetum persists throughout anther development in *ms Allium cepa, Beta vulagaris,* left flower of the spikelet of *Triticum aestivum* having *Ae. ovata* and *caudata cytoplasm.* Intact tapetal cells in the post-meiotic stages turn out to be detrimental to the growth of microspores.

3. Tapetal cells become hypertrophied in *ms Allium cepa, Capsicum annuum, Solanum melongena* and *ims* plants of *Capsicum annuum, Datura alba* was sprayed with 0·4% and 0·5% maleic hydrazide and *Sesamum indicum* treated with 0·5% dalapon and 1% maleic hydrazide. In all these cases, the tapetal cells, not only remain intact but also become hypertrophied and crush the microspores.

4. Tapetal pseudoperiplasmodium forms at various stages of anther development in the right flower of the spikelet of *ms Triticum aestivum* having *Ae. ovata* and *caudata cytoplasm, Ranunculus scleratus* plants treated with 0·2% dalapon and 0·75% maleic hydrazide and *Sesamum indicum* plants sprayed with 0·5% FW–450. Periplasmodium developed in the anthers of these plants seems to hinder the development of normal pollen grains.

Histochemical studies on localization of total carbohydrates of insoluble polysaccharides (PAS test), total proteins (Ninhydrin–Schiff's reaction), histones (Alkaline-Fast green test), DNA (Feulgen reaction) and enzyme acid phosphatase (Lead-sulphide method)[1] have shown that all the parts of an anther including tapetum in *ms* and *ims* plants are markedly deficient of these substances. It is likely that precursors, enzymes or nucleotides needed for the synthesis of these substances are absent in the anthers of *ms* and *ims* plants. This is corroborated by the fact that in all the *ms* and *ims* plants studied, the pro-cambial strand of the connective region, either fails to differentiate into vascular tissue or develops only into a limited number of thin-walled, narrow xylem elements with degenerated phloem tissue (Figs. 1–3). It therefore, seems logical that the nutrients needed for proper development of microspores which pass through tapetum[2], are not available. Tapetum showing diverse forms of abnormal behaviour is poor in metabolic activity due to starvation. The connective parenchyma also shows signs of degeneration.

The present study also reveals another notable feature in *ms* and *ims* plants of *Beta vulgaris, Capsicum annuum, Datura alba* and *Solanum melongena* (Figs. 1–4). Hyaline intracellular granules of various shapes and sizes appear in the connective parenchema. Similar types of granules have been observed in various plant parts infected by viruses[3].



FIGS. 1–4. T.S. anther connective of *ms* and *ims* plants showing hyaline intra-cellular granules in the connective parenchyama and poorly differentiated vascular tissue. 240 ×. HG ; hyaline granules ; VT : vascular tissue. Fig. 1. *ims Capsicum annuum,* Fig. 2. *ms Beta vulgaris,* Fig. 3. *ms Solanum melongena,* Fig. 4. *ms Capsicum annuum.*

Recently reduction in pollen fertility has also been observed in plants infected by viruses[4]. No inference, can however, be drawn from this excepting that these intra-cellular inclusions may be the result of some response to depletion in vascular supply in *ms* and *ims* anthers through internal causes whereas in virus infected plants, the agency may be the viruses themselves.

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R.B.S. College, Agra,
July 18, 1974.

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1. Jensen, W. A., *Botanical Histochemistry,* W. H. Freeman and Co., San Francisco, 1962, p. 408.
2. Maheshwari, P., *An Introduction to the Embryology of Angiosperms,* McGraw-Hill Book Co., Inc., London, 1950, p. 453.
3. Smith, K. M., *A Text Book of Plant Virus Diseases,* J. and A. Churchill Ltd., London, 1957, p. 652.
4. Laser, K. D. and Lersten, N. R., *Bot. Rev.,* 1972, 38 (3), 425.

SHORT SCIENTIFIC NOTES

Thiazine Dyes as Indicators in Titrations with Iodine

Out of the thiazine group of dyes, methylene blue has been used as iodometric indicator in the titration of sodium thiosulphate and arsenic (III)[1]. We now report the use of six other thiazine dyes Thionine, Methylene Green, New Methylene Blue, Toluidine Blue (Colour Index Nos. 52000, 52020, 52030 and 52040 respectively), Azure A and Azure C as indicators in the iodometric titration of sodium thiosulphate and arsenic (III). The proposed indicators have the following advantages over starch : (1) solubility in water and (2) tolerance to the presence of sodium chloride and alcohol.

Solutions 0·01% of the dyes were prepared in deionized water. Thionine and Azure C used in this investigation were Riedel-Dehaenag Seelza-Hannover and Chroma respectively, while the rest of the dyes used were Gurr's samples. Standard solutions of sodium thiosulphate, arsenic (III) and iodine were prepared as described by vogel[2].

Recommended Procedure : An aliquot of sodium thiosulphate or arsenic trioxide was taken in a 250 ml. iodine flask and diluted to 50 ml. 0·3 ml. of the solution of the dye was added (in the case of methylene green, 0·5 ml. of the solution) and the solution was titrated with iodine. The end point was from blue to green, (Purple blue to gray in case of thionine), and very sharp. While titrating arsenic (III) with iodine 0·5–1 gm. of sodium bicarbonate should be added to the titrant system. The reverse titration of iodine with thiosulphate or arsenic (III) can be carried out under the same conditions as the direct titration. The end point in this case was from green to blue (in all cases except in the case of thionine, where it is gray to purple blue). Titrations in dilute solutions (0·01 M) are also possible with a sharp end point with a blank correction of 0·06 ml. of the titrant for 0·3 ml. of the indicator. The indicator action was attributed to the formation of hydroiodide of tetraiodo dye as was suggested by Gautier[1], for methylene blue.

Two of us (N. S. N. P. and G. V. R.) thank the Council of Scientific and Industrial Research, for the award of Junior Research Fellowships.

Department of Chemistry, Andhra University, Waltair 530 003, *August* 12, 1975.

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N. S. N. Prasad.
G. V. Ramanadham.

1. Gautier, J. A., *Ann. Pharm. Franc.*, 1948, 6, 171.
2. Vogel, A. I., *A Text Book of Quantitative Inorganic Analysis*, Third Edition, The English Language Book Society, 1969, p. 349.

A Preliminary Observation on Cannibalistic Habit in Lesser Grain Borer, *Rhizopertha dominica* Fabr. (Coleoptera : Bostrychidae)

Cannibalistic habit in Red Flour Beetle, *Tribolium castenium* Herbst. has been reported by Lloyed (1957) and egg cannibalism by Sonleither (1961). But nothing is known about this habit in the lesser grain borer, *Rhizopertha dominica* Fabr. The present writers have observed this habit in *R. dominica* Fabr., for the first time.

Adult beetles were observed attacking disabled members in the rearing jars at 27 ± 1° C with 70% R.H. and eating them. However the entire body was not eaten. This habit was more pronounced in mature adults than in newly hatched ones. They aggressively attack the disabled individuals and consume their antennae, pronotal shield and abdomen. It was also observed that when *R. dominica* were reared at high temperatures (32° C with 60–70% R.H.) the cannibalism was much more pronounced even when abundant food was available. Cannibalism is not noticed under sparsely populated conditions. Only under crowded conditions and high temperature, the habit is pronounced. Cannibalism slows down below 25° C even under crowded conditions. This leads us to believe that high temperature could be an influencing factor.

On the other hand, temperature as an influencing factor has not been cited in cannibalism in *Tribolium*. Observations on *Rhizopertha dominica* Fabr. show that high temperature coupled with overcrowding leads to cannibalism.

Sincere appreciation is expressed to Professor S. M. Alam, for providing necessary facilities and to Messrs. Shamshad Ali and Masood A. Zuberi, for their assistance in the work.

Zoological Laboratories, Aligarh Muslim Univ., Aligarh, India, *September* 6, 1975.

S. Kamal.
A. Razvi.
M. Mushtaque Ahmad.
Mehr-e-Alam Khan.

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1. Lloyed, M., *Ph.D. Thesis*, Univ. Chicago, 1957, 3, 124.
2. Sonleitner, F. J., *Physiol. Zool.*, 1961, 34, 233.

Lolium remotum Schrank var. *aristatum* (Doell) Aschers (Poaceae)—A New Record for India

While making a study of the genus *Lolium* Linn. occurring in India the author came across some interesting specimens. Collections from Himachal Pradesh and Uttar Pradesh had been annotated as *Lolium temulentum* Linn. After a critical examination of the specimens they were identified as *Lolium remotum* Schrank var. *aristatum* (Doell) Aschers. Bor (1960, p. 545), had stated in the foot-note thus : "The variety *aristatum* has not so far been found in India". No other worker had reported it so far from India.

Lolium remotum Schrank var. *aristatum* (Doell) Aschers in Fl. Brand. 1864, 1 : 876. Based on *L. Linicolum* var. *aristatum* Doell, 1857. An annual, short grass ; lemmas 4·5–5·5. mm long, elliptic to ovate, turgid at maturity, awned, awns up to 1 cm. long. terminal, weak, scabrid.

Flowering and fruiting : April–September.

Distribution : Germany ; India : Himachal Pradesh, Uttar Pradesh.

Exsicc. : Himachal Pradesh–Dalhousie, 6000 ft., 17th September, 1874, *C.B Clarke* 22611 (CAL ; Uttar Pradesh–Dehra Dun Dist., 30th April 1969, *H.B. Naithani* 6568 (DD).

I am thankful to the Director, Botanical Survey of India, Calcutta, for facilities extended, and to Dr. R. B. Majumdar, Botanical Survey of India, for giving suggestions.

Central National Herbarium,
Shibpore, Howarh 711 103,
August 26, 1975.

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1. Bor, N. L., *The Grasses of Burma, Ceylon, India and Pakistan (Excluding Bambuseae)*, Pergaman Press, 1960, p. 545.
2. Turrell, E. E., "Taxonomic revision of the genus *Lolium*," *Tech. Bull.*, 1968, 1392, 38.

A New Leaf Spot of Wheat Caused by *Chaetomium* Sp.

During September, 1974, while making disease observations on Plant Pathological Screening Nursery (PPSN) at Keylong (Lahaul and Spiti Valley of Himachal Pradesh) about 8,000 f. a.s.l., one entry was observed to be severely attacked with leaf spot sympotoms. These spots were elliptical, irregular, straw coloured and were more pronounced on matured leaves. Isolations were made from the surface sterilized diseased spots on 2% potato-dextrose-agar (PDA). These isolations yielded a species of *Chaetomium*. The culture was purified by single ascospore isolations and was maintained on PDA. On the basis of cultural characters the fungus was identified as *Chaetomium dolichotrichum* (Ames, 1945 and 1961).

The pathogenicity tests were conducted on one month old plants of wheat variety Bolley golden × W, 1929, by inoculating with spore and mycelial suspension. Six or seven days after inoculations the disease symptoms appeared as small, irregular, elliptical, light brown coloured spots on the lower leaves of the inoculated plants, the uninoculated plants remained free. Reisolations from these spots yielded the same fungus. The present communication is the first record of *C. dolichotrichum* causing leaf spots disease in wheat.

Its culture has been deposited in the Indian type culture collection, I.A.R.I., New Delhi, under the accession Number 1949.

The authors are thankful to Dr. J. N. Kapur, Mycologist, Indian Agricultural Research Institute, New Delhi, for the identification of the fungus.

Division of Mycology and Plant Pathology,
Indian Agricultural Research Institute, New Delhi,
November 17, 1975.

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K. D. SRIVASTAVA.

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1. Ames, L. M., *Mycologia*, 1945, 37, 145.
2. —, *A Monograph on Chaetomiaceae*, 1961.

ANNOUNCEMENTS

Award of Research Degrees

Tamil Nadu Agricultural University has awarded the Ph.D. degree in Agriculture to Shri R. Palaniappan and Shri A. Dhanapalan Mosi; Shri P. Kandaswamy, Shri Suryanarayana Veera, and Shri G. Arunachalam.

Annamalai University has awarded the Ph.D. degree in Marine Biology to Shri K. V. Venkataramanujam.

Osmania University, Hyderabad, has awarded the Ph.D. Degree in Geology to Shri Karri Sivaji ; Ph.D. degree in Zoology to Smt. Suraiya Rasheed ; Ph.D. degree in Biochemistry to Shri Raja Mohan K.

Utkal University, Bhubaneswar, has awarded the Ph.D. degree in Mathematics to Shri Jugal Charan Muduli ; and Shri Dola Gobinda Sahoo.

Karnatak University, Dharwar, has awarded the Ph.D. degree in Physics to Shri M. B. Shankar ; Ph.D. degree in Botany to Shri S. P. Hosmani ; Ph.D. degree in Zoology to Shri S. B. Deshpande.

Sri Venkateswara University, Tirupati, has awarded the Ph.D. degree in Physics to Shri B. C. Venkata Reddy, Ph.D. degree in Statistics to Shri V. Lakshminarasimham ; Ph.D. degree in Mathematics to Shri D. Bathaiah ; Ph.D. degree in Chemistry to Shri C. Devendranath Reddy ; Ph.D. degree in Zoology to Shri N. Chandrasekharam Naidu.

REVIEWS AND NOTICES OF BOOKS

Annual Review of Entomology (Vol. 20), Edited by Ray F. Smith, Thomas E. Mittler and Carroll N. Smith. (Annual Reviews, Inc., Palo Alto, California 94306 USA), 1975. Pp. 514. Price: USA $ 12.00, Foreign $ 15.50.

The Annual Review of Entomology—Volume 20, 1975 is out. It contains twenty-one valuable titles duly writtern by eminent specialists of their respective subjects. The articles on various aspects of biological, ecological, physiological, taxonomical, applied and legislative fields of insect and other pests are covered therein. Apart from the author and subject indexes, cumulative index of contributing authors alongwith their chapter titles from volumes 11 to 20 in a well arranged manner are also given at the end of this book for the convenience of readers.

The topics of the articles contained in this volume are: Biology and Control of Imported Fire Ants; Biological Control of Aquatic Weeds; Biology and Ecology of Armored Scales; Physiology of Tree Resistance to Insects; Status of Viruses Pathogenic for Insects and Mites; Federal and State Pesticide Regulations and Legislation; Neurosecretion and the Control of Visceral Organs in Insects, Neuromuscular .Pharmacology of Insects; Inherited Sterility in Lepidoptera; Evolution and Classification of the Lepidoptera; Recent Developments in Insect Steroid Metabolism; Recent Research Advances on the European Corn Borer in North America; Recent Advances in our Knowledge of the Order Siphonaptera; Adaptations of Arthropoda to Arid Environments; Responses of Arthropod Natural Enemies to Insecticides; Plant Resistance to Insects Attacking Cereals; Brain Structure and Behaviour in Insects; Structure of Cuticular Mechanoreceptors of Arthropods; The Brazilian Bee Problem; Insect Growth Regulators with Juvenile Hormone Activity and Genetical Methods of Pest Control.

I have gone through all the articles and each review is of standard value having due importance in its respective branch of Entomology. Special emphases of the readers are drawn to the reviewed articles on Plant Resistance to Insects Attacking Cereals by R. L. Gallun *et al.*, and Genetical Methods of Pest Control by M. J. Whitten and G. G. Foster which are of applied nature from the Economic Entomology point of view. In the last mentioned article, the authors have reviewed the developments in the Sterile Insect Release Method (SIRM) subsequent to Proverbs' comprehensive treatment in Volume 14 of *Annual Review of Entomology* (49). They have described more ambitious goals of genetic control and have discussed their future prospects. Various examples where success have been achieved are cited. Genetic manipulation methods to reduce the size of the pest population or removing the noxious characteristic of pest are also described.

This valuable volume is difficult to be purchased by every individual; but must be purchased for each library of the Research Institutes and Colleges for the benefit of students and research workers.

V. G. Prasad.

Die Saugotiere der Sowejet-Union. Band II. By Seekühe und Raubtiere. (Gustov Fisher Verlag Jena), 1974. Pp. 1006, Price 239.40 M.

This German translation of the original Russian edition published in 1967 deals with 30 species of seacows, wolves, foxes, wild dogs, bears, martens, weasels, otters and minks of the Soviet Union. This exhaustive account runs over 1000 pages of text, and the book constitutes a veritable treasure-house of information on taxonomy, geographical distribution, habitat preferences, daily and seasonal movements, food and feeding habits, breeding cycles, population dynamics and social behaviour of the species involved.

Madhav Gadgil.

The Grass Cover of India. By P. M. Dabadhao and K. A. Shankaranarayan. (Indian Council of Agricultural Research, New Delhi), 1973. Pp. xii + 1–713. Price Rs. 35·00. 18·5 × 24 cm.

A large country like India has agriculture as the main stay and in view of solving the food problem for a teaming population of this country, it is essential that more and more factual information is brought out on different aspects of agriculture. From this point of view and because of the fact that the cattle wealth of India is relatively large, the need for knowing accurate information on the grass cover of India is all the more imperative at a time when agriculture has been rapidly progressing in our country and more so since grass forms the basic food material for cattle, sheep and other ungulates.

After a brief preface, this extensive treatise is dealt under 2 parts. The first part which covers 197 pages relates to types of grass covers, in relation to environment and management. Such aspects as environment, natural vegetation and grass cover, review of earlier literature, techniques adopted in grassland survey of India, types of grass covers, grass communities of the states, ecological characteristics of important species of grasses, management and conservation of the grass cover and finally an exhaustive bibliography consisting of 202 references are dealt with in a cogent manner and the information provided is very exhaustive. A redeeming feature of this part is the inclusion of 16 maps covering various aspects in relation to the several chapters covered here. These aspects are particularly interesting because they relate to grassland ecology from the point of view of its topography and geomorphology, climate, soils and biotic factors including activities of man and animals who are the causal agents for deforestation, shifting cultivation, grazing, fire and other such associated factors.

Continuing further, 5 major types of grass covers are recognised and they are the following: *Schima-Dicanthium* Type; *Dicanthium-Cenchrus-Lasiurus* Type; *Phragmites-Saccharum-Imperata* Type; *Themeda-Arundinella* Type and Temperate-Alpine Type. While describing these types of covers in detail, very useful notes on floristic composition, distribution, factors on habitat and succession of the principal species of grasses are provided. The last chapter of this part is particularly interesting because it points out the need for research in grassland ecology and improvement in management.

The second part on grassland survey of India reports on an analysis of 500 sites in 19 States of India. For each of these States, the number of sites studied is provided; thus for Andhra Pradesh (Sites 1–28), Assam (Sites 29–40) and so on. In these reports, some of the important aspects which are dealt in an adequate manner are on the location of the sites, soil geomorphology, plant litter, total plant cover, forage, floristic composition, percentage of frequency of grass, percentage cover of forest type and subtypes. If references to authentic herbarium material on which the reports on the floristic composition of the grass cover are based were provided, it would have been more useful and valuable.

For both the parts, two separate indices are provided for plant names. On the whole, the authors have taken a lot of trouble in amassing a wealth of information. Hence, both the authors and the ICAR have to be congratulated because this treatise on the grass cover of India will definitely serve as a very valuable guide for ecologists, agrostologists, applied scientists, and those interested in soil conservation and grassland husbandry in relation to general improvement of livestock. Without any hesitation, this commendable work can serve as a very important reference work on grassland in particular and its bearing on the improvement of cattle in general.

K. SUBRAMANYAM.

Index of Vibrational Spectra of Inorganic and Organo Metalic Compounds. (Vol. II). By N. N. Greenwood and E. F. G. Ross. (Butterworths, London), 1975. Pp. vii + 908. Price £ 40.00.

This is the second volume in the series covering the literature pertaining to the vibrational spectra of inorganic and organometallic compounds. The literature covered is through 1961–63. The book provides an index of numerous compounds whose vibrational spectra (either Raman or Infrared) are available.

In the introductory chapter, the authors have outlined the scope of the survey of literature. In their criteria, compounds are considered organic if they contain carbon and hydrogen with As, Br, Cl, F, I, N, O, P, S or Se, as the only other elements or if they contain only the elements As, Br, C, Cl, F, I, N, O, P, S or Se and in addition a C–C bond. Also ionic compounds, containing an organic anion and mono atomic cation is excluded if the cation is Na^+, K^+, Rc^+, Cs^+, Fr^+, Ca^{2+}, Sr^{2+} or Ba^{2+}. This eventually answers the non-inclusion of certain compounds. The table consists of information regarding the compound, state in which the spectrum is taken, type of the spectrum (Raman or infrared), range in wave numbers and the corresponding reference.

This is a faithful representation of literature in a form necessary for the various workers who need this information. There exist other books in this area written by Nakamato, Adams, Ross, Szyamanski and others, and data books compiling information regarding the spectra. The aim of this book, as I realise, is for data dissemination; however, I am skeptical as to the utility of such books for intended users. This comment does not mean to reflect the credibility of the authors but the basic theme on which this publication is conceived.

V. KRISHNAN.

5737/76. Published by S. R. S. Sastry, Manager, Current Science Association, Bangalore-560006
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INFORMATION TO CONTRIBUTORS

CURRENT SCIENCE is the Premier Science Fortnightly of India published by the Current Science Association, Bangalore and issued on the 5th and 20th of each month. All material intended for publication in CURRENT SCIENCE and books for review should be addressed to the Editor, *Current Science,* Raman Research Institute, Bangalore-560006.

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Articles and papers should include a short abstract at the beginning.

References are indicated by superscripts in the text. The style of references should be

Journal: The names of the authors, the journal, year, volume and page.

Book: Authors names, the title of the book, name and location of the publishers, year and page.

"Personal Communications" and "Unpublished Work" do not go into references and should be incorporated in the text. Acknowledgements should be brief.

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SPIN AND TORSION PROVIDE THE BOUNCING MECHANISM FOR A NON-SINGULAR PULSATING UNIVERSE

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IN the beginnings of general relativistic cosmology, the concept of an oscillating universe, with alternating cycles of expansion and costraction, has been popular. The Hawking–Penrose singularity theorenms[1], have pointed to the inadequacy of the concept of a pulsating universe within the standard framework of general relativity (with zero cosmological constant) : Already for a single expansion-contraction cycle, there are two barriers (one at a finite time in the past, another at a finite time in the future) beyond which the space-time manifold cannot be extended. The well-known trouble with the cosmological singularity is doubled for closed models.

Of course, the singularity does not mean any breakdown of physics. Only classical physics stops here, and it is often conjectured that with some quantum theory of gravity, replacing the classical formulation of general relativity at some sufficiently contracted stage of matter evolution, it should be possible to pass in a smooth manner from a stage of contraction to a subsequent of expansion. The singularity should be conceived thus only as in unrealistic consequence of the extrapolation of the classical general relativity theory to situations where it evidently cannot apply.

We have, so far, no consistent quantum formulation of the general theory of relativity. But when a characteristic physical quantity of the quantum theories—the spin, has been incorporated as a dynamical quantity into the general relativistic theory of space-time, this proved to be a sufficient step toward the expulsion of the unphysical, singularities from cosmology. This "general relativity with spin" is called the Einstein–Cartan theory of gravitation, briefly ECT[2,3]. The geometric structure of the space-time manifold is generalized in it from a purely Riemannian one into a general affine one, with non-vanishing torsion which is algebraically related to the density of the spin angular momentum of matter. Torsion is identically zero where matter density is zero, thus equations in general relativity and in the ECT are the same for empty space ; at the present observational level, these two theories are practically indistinguishable.

The characteristic spin-spin interaction of matter in the ECT which dominates the behaviour of matter of extremely high densities, above ca. 10^{50} g cm^{-3}, is able to prevent singularities. With the simple model of a Weyssenhoff fluid[4] of energy density ρ, pressure p, and spin density vector S_i (orthogonal to the four-velocity u_i in a comoving frame of reference), one of the two alternative approaches may be adopted : Either (I) the spin vector S_i conforms to the symmetries of the space-time under study, *i.e.,* the spins of matter are aligned along some characteristic direction of anisotropy, or (II) the spins are randomly distributed, and in averaging over them only some spin-square terms remain.

Cosmological models with aligned spins have to be at least partially anisotropic ; extensions of the isotropic Robertson–Walker models to the ECT are thus impossible. The study of aligned spin models started two years ago with the Bianchi type I[5]. A lot of exact solutions and various results derived in the meantime are recently summarized[6]. More than two years ago, Trautman[7] thought it interesting to know whether a closed model would also bounce because of spin and torsion. The possibility of such a bounce, for aligned spin models, has not been proved till now for the general Bianchi type IX models which are a generalization of the closed Robertson–Walker cosmologies. But it has been proved very recently[8], that pulsating models with a 4-parameter. Lie group of isometry which does not permit any simply transitive group of motions are possible. These models constitute non-singular ECT analogues of the three-cylinder semiclosed cosmological models of Kantowski and Sachs[9]. In contrast to the closed Robertson–Walker models, they are always shearing. In ECT, the main scale-factor R for them is always $R \geqslant R_{min} > 0$ provided a certain condition holds[8] which expresses the fact that the effect of the spin term should overwhelm the effect of the shear when $R \rightarrow R_{min}$. This condition for the prevention of the singularity looks much like analogous conditions for Bianchi type I models[5,6].

Two other questions raised by Trautman[7], have been solved in the meantime: the effect of the pressure, and of the magnetic field H which should be responsible for the alignment of spins. When energy density ρ and pressure p are related by a linear equation of state : $p = \gamma\rho$, with $0 \leqslant \gamma < 1$, then non-singular ECT solutions of several Bianchi types already exist[10]. Also the pulsating semiclosed models of Kantowski–Sachs type allow for non-zero pressure, while being non-singular in the ECT. Finally, exact non-singular cosmological models of axial symmetry, with a non-vanishing magnetic field pointing in the direction of spin alignment, have been constructed[11]. The ECT allows even for static models containing a uniform magnetic field and spinning matter which were discussed earlier in this journal[12].

With random spin models, the situation is even simpler than for models with aligned spins. Since spins may locally fluctuate in direction, and only averages are meaningful, there are now no troubles with adapting the spinning Weyssenhoff fluid to the high symmetries of the Robertson-Walker models. Closed, flat and open Robertson-Walker models with random spin distribution are possible[13]. Below we give the simple expression for the radius function R of a pulsating Robertson-Walker model, filled with spinning dust of an averaged spin square density $< S^2 > = S_0^2/R^4$. This is : $2R - D = (D^2 - S_0^2) \sin D/2 \, (t + \sqrt{DR - R^2 - \frac{1}{4} S_0^2})$, where D is some integration constant $(D^2 > S_0^2)$, and t is the cosmic time. This model pulsates between the minimum radius $\frac{1}{2} (D - \sqrt{D^2 - S_0^2})$ and the maximum radius $\frac{1}{2} (D + \sqrt{D^2 - S_0^2})$. A characteristic difference with respect to the situation in general relativity (where $S_0 \equiv 0$) occurs : The spin-spin interaction slightly raises the minimum radius from zero and at the same time it decreases also slightly the maximum radius from its general relativistic value.

We gave here this exact solution for a non-singular pulsating universe with randomized spins merely as an example ; other related solutions are being derived and will be published together elsewhere. Our aim is to underline here that the possibilities of random spin models in the ECT are even higher than those of aligned spin models, both with respect to variety of types and to the possibility of incorporating the spinning matter in an already given Bianchi (or another) type. Thus Bianchi type II non-tilted models do not allow for a generalization into ECT models[10], even into singular ones, as the spin distribution is aligned, *i.e.*, it conforms to the symmetries of this type. But these, as well as *all* known models of general relativity may be incorporated into the ECT as models with randomly oriented spins, and in most cases (till now I could find no counterexample) they may be rendered free from singularity by the spin-spin interaction. Since the time when Trautman[7] asked whether a bounce of closed models because of torsion is possible, there accumulated thus much positive evidence in favour of this bounce. The only dubious case is that of Bianchi type IX with aligned spins.

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1. Hawking, S. W. and Ellis, G. F. R., *The Large Scale Structure of Space-Time*, Cambridge University Press, Cambridge, 1973.
2. Hehl, F. W., *Gen. Rel. and Gravit.*, 1973, 4, 333 ; 1974, 5, 491.
3. Trautman, A., *Istit. Nazion. Alta Matem.*, *Symposia Matematica*, 1973, 12, 139 ; *Bull. Acad. Polon., Sér. Sci., Math., Astr., Phys.*, 1972, 20, 185, 503, 895.
4. Weyssenhoff, J. and Raabe, A., *Acta Phys. Polon.*, 1947, 9, 7.
5. Kopczyński, W., *Phys. Letters*, 1973, **43 A**, 63.
6. Kuchowicz, B., *Acta Cosmologica* (Kraków), 1976, 4, in press.
7. Trautman, A., *Nature Phys. Sci.*, 1973, 242, 7.
8. Kuchowicz, B., *Phys. Letters*, 1975, **54 A**, 13.
9. Kantowski, R. and Sachs, R. K., *J. Mathem. Physics*, 1966, 7, 443.
10. Tafel, J., *Acta Phys. Polon.*, 1975, **6 B**, 537.
11. Kuchowicz, B., *Astrophysics Space Science*, 1975, 38, in press.
12. —, *Curr. Sci.*, 1975, 44, 537.
13. —, *Cosmology Without Singularity*, Essay, selected for "Honorable Mention," at the Gravity Res. Found. Essay Competition, 1975.

RAPID ESTIMATION OF LYSINE BY HIGH VOLTAGE ELECTROPHORESIS

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ABSTRACT

High voltage electrophoresis technique has been adopted for rapid estimation of lysine. Pyridine : acetic acid water (100 : 10 : 800 v/v), pH 6·2, gives excellent separation of lysine, argioine, histidine, glutamle acid and aspartic acid. - Each separation takes 20–30 min. at 3,000–4,000 volts. The values obtained agreed very closely (r-0·0967) with those obtained by the manometric method.

CONSIDERABLE progress has been made in rapid estimation of protein by different techniques. Since lysine happens to be the first limiting amino acid in almost all the cereal grains[1-3], its estimation is quite important in the improvement of the protein quality and in the development of varieties with higher nutritive value. At present lysine is mostly estimated either by employing autoanalyser[4] or by Warburg technique[5]. In both the cases lysine is decarboxylated and the resultant carbondioxide produced is estimated either manometrically or colorimetrically.

When a large number of samples has to be analysed, these two methods cannot be used as screening techniques. The first one requires expensive equipment and the second method is time consuming and enzyme dependent. Colorimetric method for lysine estimation has also been reported[6]. In the present study high voltage electrophoresis has been adopted for the rapid estimation of lysine. In this procedure it is not necessary to completely remove the hydrochloric acid after hydrolysis and the acid can be neutralised by the addition of alkali after the first evaporation. This saves considerable amount of time. The pyridine : acetic acid : water buffer system. (pH : 6·2) gives good resolution for lysine, arginine, histidine, glutamic acid and aspartic acid and all these amino acids can be estimated. Values for lysine as estimated by high voltage electrophoresis and Warburg technique agreed closely.

EXPERIMENTAL

A weighed sample (250 mg) was mixed with 8 ml 6 M HCl in hydrolysing tubes and sealed after evacuation. Hydrolysis of protein was done at 110° C for 18 hr. Hydrolysate was filtered and the filtrate collected was evaporated and the residue was dissolved in 1 ml buffer and neutralised NaOH (6 N).

The chromatographic paper (20 × 40 cm) was impregnated with pyridine : acetic acid : water (100 : 10 : 800 v/v) pH 6·2 and the excess buffer was removed, 5 μl of the sample was applied at the centre. Electrophoresis was carried out using Camag High Voltage Electrophoresis equipment at 3000–4000 volts for 20–30 minutes depending on the voltage applied. After this the paper was removed, allowed to dry in air and sprayed with ninhydrin (0·2% in ethanol). The colour was developed at 110° C for 2 minutes. The spots were cut and eluted with 70% acetone and read at 590 nm. The quantity of lysine was calculated from a standard curve prepared in the same way. Lysine was also estimated by conventional Warburg technique[5].

RESULTS AND DISCUSSION

Of the different buffer systems tried pyridine : acetic acid : water (100 : 10 : 800 v/v) pH 6·2 gave good separation and resolution for the five amino acids, lysine, arginine, histidine glutamic acid and aspartic acid (Fig. 1). Sample was applied in the centre of the paper. Lysine, arginine and histidine migrated towards the cathode while glutamic acid and aspartic acid migrated towards anode. The other amino acids remained very near the origin where the sample was applied. In this estimation the pH of the buffer is quite critical as the lower pH was found to separate other amino acids as well but the separation of lysine from arginine and histidine was not good. At 4000 volts each separation took 20 min. only and three samples in duplicate were applied each time.

The amount of lysine obtained in 19 different samples representing maize endosperm, embryo and pericarp by this technique as well as conventional Warburg technique is shown in Table I. It is seen that the amount of lysine obtained by high voltage electrophoresis is in very close agreement with the amount obtained by Warburg technique. The correlation coefficient value was 0·987. High voltage electrophoresis technique is much faster than the Warburg technique and complete removal of the acid after hydrolysis is not necessary and acid can be neutralised with alkali after first evaporation thereby saving considerable time.

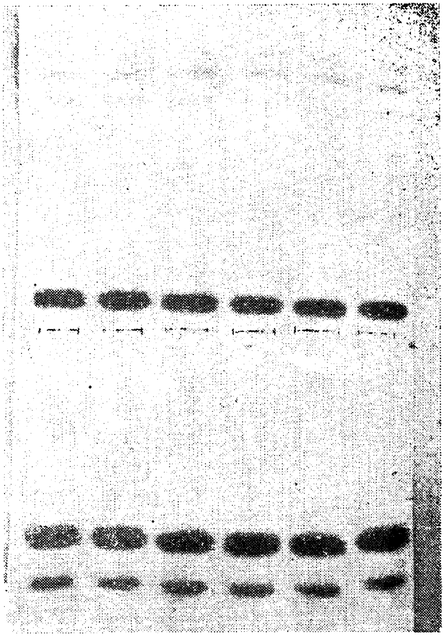

FIG. 1. Separation of amino acids by High Voltage electrophoresis. Top to bottom: Lysine, arginine, histidine, neutral amino acids, glutamic acid and aspartic acid. Line at the centre represents origin where the samples were applied.

Besides the estimation of lysine, other amino acids, namely, arginine, histidine, glutamic acid and aspartic acid can also be estimated. This is not possible with either Warburg technique[5] or colorimetric estimation for lysine[4-6]. Since this method is rapid, it has a potential to be used as a screening technique for estimation of lysine.

TABLE I

Lysine content in g/100 g as determined by High Voltage Electrophoresis and Warburg technique

Sample No.	High Voltage Electrophoresis Method	Warburg Method
Opaque-2 kernel	0·34	0·31
,,	0·32	0·32
,,	0·36	0·37
,,	0·40	0·39
,,	0·39	0·40
,,	0·38	0·40
,,	0·34	0·33
,,	0·32	0·38
Pericarp	0·12	0·12
Opaque-2 Percarp	0·19	0·22
Percarp	0·16	0·16
Endosperm	0·32	0·34
Embryo	0·48	0·50
Endosperm	0·29	0·33
Embryo	0·58	0·59
Opaque-2 Endosperm	0·34	0·36
Opaque-2 Embryo	0·50	0·48
Endosperm	0·30	0·29
Embryo	0·59	0·61

Correlation Coefficient — 0·987**

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1. Carpenter, K. J., *Nutr. Soc. Eng. Scolt.*, 1958, **17**, 91.
2. Martinez, W. H., Frampton, V. L. and Cabell, C. A., *J. Agr. Food Chem.*, 1961, **9**, 64.
3. Nelson, O. E., *Adv. Agron.*, 1969, **21**, 171.
4. Schaiberger, G. E., *Ann. N.Y. Acad. Sci.*, 1960, **87**, 890.
5. Umbreit, W. W., Burris, R. H. and Stauffer, J. E., *Manometric Techniques*, *Burgess, Minneapolis, Minn.*, 1957.
6. Tsai, C. Y., Hansel, L. W. and Nelson, O. E., *Cereal Chem.*, 1972, **49**, 572.

INSTITUTION OF CHEMISTS (INDIA) ASSOCIATESHIP EXAMINATION, 1977

The twenty-seventh Associateship Examination of the Institution of Chemists (India) will be held in November, 1977. The last date for Registration is 30th November, 1976. The Examination is recognised by the Government of India as equivalent to M.Sc. in Chemistry, for purposes of recruitment of Chemists. Further enquiries may be made to the Honorary Secretary, Institution of Chemists (India), Chemical Department, Medical College, Calcutta-73.

SOME BIOCHEMICAL CHARACTERISTICS OF MODIFIED PHENOTYPE STRAINS OF OPAQUE-2 (*ZEA MAYS* L.)

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ABSTRACT

Studies with modified strains of opaque-2 maize with vitreous endosperm fractions showed that their endosperm generally contained more protein and less tryptophan compared to chalky-opaque endosperm types. Increase in kernel vitreosity (opaque→ 100% vitreous) was associated with an increase in the protein and a decrease in lysine and tryptophan in endosperm. The vitreous fraction of the endosperm contained more protein but less tryptophan than the opaque fraction.

NUTRITIONAL quality of protein in normal maize is relatively low since it is deficient in the essential amino acids lysine and tryptophan. The discovery by Mertz, Bates and Nelson[1], that the maize mutant opaque-2 has nearly twice as much lysine and tryptophan as normal maize, opened up new vistas in improving cereal protein quality. Opaque-2 maize varieties have in recent years been developed and recommended for cultivation in several countries[2]. Although the nutritional superiority of opaque-2 maize over ordinary maize has been well established in experiments with rats[3], swine[4], young childern[5,6], and adult humans[7]; yet it has shown limited acceptability among the farmers because of some agronomic and acceptability problems[2]. These problems are associated with soft endosperm and dull chalky appearence of opaque-2 maize kernels. Efforts were made to develop modified strains of opaque-2 maize[8] with vitreous (normal) endosperm fraction. This paper reports some of the biochemical characteristics of such strains.

Modified opaque-2 maize strains were developed from opaque-2 composites Shakti, Rattan and Proteins as described by Singh *et al.*[8]. After harvest, opaque and various categories of modified opaque-2 kernels were separated on the basis of their vitreousness by screening them against light. For comparing opaque and vitreous fractions of modified opaque-2 kernels (½ normal–½ opaque), a sample of modified endosperms from each family was separated into opaque and vitreous fractions. Twenty randomly selected kernels of each type were taken and analysis was made on defatted endosperm samples. Protein was estimated by micro-Kjeldahl procedure[9], tryptophan by colorimetric method[10] using papain enzyme for hydrolysis and lysine by Amino acid Analyser.

Results presented in Table I showed that modified opaque-2 kernels, in general, contained more protein in the endosperm than opaque kernels. For ear Nos. 181 and 464, however, there was small decline in protein. On the other hand, the per cent tryptophan in the endosperm protein in the modified kernels was generally lower than chalky-opaque kernels. However, ear Nos. 353, 409, 464 and 508 were most desirable since there was little change in tryptophan content. These results suggested the possibility of selecting modified opaque-2 kernels having tryptophan in protein comparable to chalky-opaque kernel types.

TABLE I

Protein and tryptophan contents of opaque and modified phenotype opaque–2 kernels selected from different ears*

Ear*	% Protein		% Tryptophan in protein	
	Opaque	Modified	Opaque	Modified
12	6·70	9·24	0·90	0·83
34	6·45	8·09	1·22	0·95
181	6·58	6·47	1·06	0·88
353	9·01	12·01	0·71	0·70
[illegible]89	6·70	8·78	1·25	0·80
399	6·81	7·16	1·13	0·93
405	10·05	12·47	0·77	0·51
409	10·05	11·09	0·77	0·76
464	9·93	9·47	0·83	0·86
508	8·55	8·78	0·79	0·75
Mean	8·08 ±0·48	9·36 ±0·59	0·94 ±0·06	0·80 ±0·04

* Analysis is based on defatted endosperm.

Studies with different categories of modified opaque-2 kernels showed that protein and amino acid composition of endosperm were affected by kernel vitreosity (Table II). With the increase in kernel vitreosity (opaque → nearly normal), protein content increased from 9·4% to 12·6% but lysine and tryptophan (in protein) decreased, respectively, from 3·6% to 1·6% and from 0·96% to 0·43%. Although from grain yield and acceptability point of view 100% vitreous kernel type would be more desirable, yet from quality and phenotypic marker view point, this will not be suitable. Therefore, a compromise has to be made. Thus semi-normals (50% opaque—50% vitreous) seem to be more desirable since they combine high levels of protein and tryptophan in hard endosperm background[11].

TABLE II

Protein, lysine and tryptophan contents of different categories of hard endosperm opaque-2 kernels selected from an opaque-2 composite*

	Phenotype	% Protein	% Lysine in protein	% Tryptophan in protein
1.	100% opaque	9·44	3·58	0·96
2.	25% normal—75% opaque	10·13	3·33	0·88
3.	50% normal—50% opaque	10·25	3·13	0·81
4.	75% normal—25% opaque	10·97	2·30	0·57
5.	More or less normal	12·06	1·63	0·43

* Analysis is based on defatted endosperm.

On comparing opaque and vitreous fractions of the endosperm of modified opaque-2 kernels, it was observed, for the families sampled, that the chalky-fraction had lower protein (per cent) but higher tryptophan (per cent of protein) than the vitreous fraction (Table III). However, the tryptophan values obtained for vitreous-fractions were lower but more reflective of values obtained for whole endosperms. These observations further supported the findings that with the increase in kernel vitreosity protein increased and tryptophan decreased.

TABLE III

Comparison of protein and tryptophan contents of opaque and vitreous portions of modified kernels (½ normal—½ opaque)

Family	Component analysed	% Protein	% Tryptophan in protein
Shakti-181	Opaque portion	7·72	0·93
	Vitreous portion	9·22	0·82
	Whole endosperm	8·94	0·37
Shakti-201	Opaque portion	6·69	1·24
	Vitreous portion	10·22	0·81
	Whole endosperm	9·19	1·02
Shakti-375	Opaque portion	7·75	1·17
	Vitreous portion	14·63	0·69
	Whole endosperm	11·69	0·73
Shakti (SO) H.S. 17	Opaque portion	7·76	1·10
	Vitreous portion	11·82	0·81
	Whole endosperm	11·04	0·92
Rattan-34	Opaque portion	6·25	1·26
	Vitreous portion	12·69	0·65
	Whole endosperm	10·75	0·75

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1. Mertz, E. T., Bates, L. S. and Nelson, O. E., *Science*, 1964, **145**, 279.
2. Singh, J. and Asnani, V. L., *High-Quality Protein Maize*, Dowden, Hutchinson and Rose, Inc., Penn, USA, 1975, p. 85.
3. Mertz, E. T., *Proc. High Lysine Corn Conf.*, Corn Refineries Association, Inc., Washington, 1966, p. 12.
4. Cromwell, G. L., Pickett, R. A., Cline, T. R. and Beeson, W. M., *J. Anim. Sci.*, 1969, **28**, 478.
5. Bressani, R., *Proc. High Lysine Corn Conf.*, Corn Refineries Association, Inc., Washington, 1966, p. 34.
6. Singh, J. and Koshy, S., *Indian J. Genet.*, 1974, **34 A**, 1182.
7. Clark, H. E., *Proc. High Lysine Corn Conf.*, Corn Refineries Association, Inc., Washington, 1966, p. 40.
8. Singh, J., Lodha, M. L. and Gupta, H. O., *Indian J. Genet.*, 1974, **34 A**, 651.
9. A.O.A.C., "Official Methods of Analysis of the Association of Official," *Agril. Chemists*, 1965, p. 744.
10. Hernandez, H. and Bates, L. S., *CIMMYT. Res. Bull.* No. 13, 1968.
11. Singh, J., Lodha, M. L. and Gupta, H. O., *Proc. VII Meeting Maize and Sorghum Section of EUCARPIA*, Zagreb–Stubicke Toplice 3–6, Sept. 1973.

NUTRITIVE VALUE OF NORMAL AND OPAQUE-2 MAIZE

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ABSTRACT

In an isonitrogenous feeding experiment with albino rats the biological value of a high lysine opaque-2 composite 'Shakti' and three normal maize varieties, namely, composite 'Vijay', 'Ganga-5' and Basi was tested. The biological value of opaque-2 maize was 90% of milk protein casein while that of normal maize varieties was less than 70% of casein. Opaqae-2 maize also had significantly higher percentage of net protein utilization and utilizable nitrogtn compared to normal maize varieties. When opaque-2 maize was fed at inherent protein level with mineral and vitamin supplementation, the weight gain in rats was 54·4 g on opaque-2 diet and 43·3 g on Balahar in a 28-day feeding trial. Feed efficiency ratios were respectively 4·01 and 4·24 for opaque-2 maize diet and Balahar.

INTRODUCTION

THE discovery opaque-2 gene results in marked increase in lysine and tryptophan contents[1], has opened up new vistas in improving protein quality. Following the work on maize, mutants with high lysine contents have also been identified in barley[2] and sorghum[3]. Most of the biochemical studies, conducted so far[4-6] have clearly established that opaque-2 maize endosperm protein contains 15–27% zein as compared to 54–59% in normal maize. Since zein is extremely deficient in lysine and tryptophan and rich in leucine, the net result is improved amino acid composition in opaque-2 maize. Besides this, normal maize protein has higher leucine-isoleucine ratio which is responsible for the development of pellegra in populations subsisting mainly on maize[7]. In the present study evaluation of protein quality of one opaque-2 composite, one normal hybrid, one normal composite and one open pollinated local maize variety has been done by nitrogen-balance study.

MATERIALS AND METHODS

Three normal maize varieties, namely, Vijay, Ganga-5 (a hybrid) and Basi (a local) and an opaque-2 composite Shakti were tested. The baby foods 'Amulspray' (milk based), and 'Balamul' (cereal and milk based), Balahar (atta, 85% and edible groundnut flour, 15% fortified with vitamins and minerals) and casein were used in this investigation.

Crude protein content (N × 6·25) was determined by micro-Kjeldahl method[8], and lysine was estimated by amino acid analyser.

Nitrogen Balance.—The N-balance of each sample was determined in five Wistar male growing rats by the Mitchell and Carman method[9]. Rats were arranged in groups of five with an average weight of 100 g. The total experimental period of nine days consisted of four days preliminary feeding followed by a five day balance period in which pooled faeces and pooled urine were analysed for nitrogen. Each rat received a constant amount of 10 g dry matter and 150 mg nitrogen daily. Water was given *ad libitum.* All grain diets and casein supplemented with 1% methionine were fed as isonitrogenous diets at 9·5% protein. Nitrogen content of each diet was adjusted using N-free diet mixture[10]. The experimental diet contained 4% mineral and 1% vitamin mixture[10]. Metabolic-N and endogenous-N were determined as described by Eggum[10].

Feed Efficiency Ratio (FER).—The FER (g of feed consumed per g of body weight gain) of Shakti, Amulspray, Balamul and Balahar at their inherent protein levels was also determined. Only opaque-2 diet was supplemented with 4% mineral and 1% vitamin mixture as other foods were already supplemented with minerals and vitamins. Wistar male growing rats having initial average weight of 41·5 g were used. Rats were arranged in groups of five. Experimental diets were fed to each rat for 28 days. Rats were given diet and water *ad libitum.* Individual weight gains and feed consumption were measured after 7, 14, 21 and 28 days. FER was calculated for each rat.

RESULTS AND DISCUSSION

Results of analysis of maize samples are given in Table I. The local maize variety Basi had the highest protein with lowest lysine content, this led to very low chemical score. The protein content of opaque-2 maize was comparable with Vijay and Ganga-5 but it had substantially higher (about 44%) lysine content. The chemical score for Shakti was highest being 40–56% greater than the normal maize varieties.

TABLE I

Chemical score of normal and opaque-2 maize varieties

Maize variety	Protein (%)	Lysine (g/16 g N)	Chemical score* (%)
Vijay	11·00	2·70	49·1
Ganga-5	11·81	2·84	51·6
Basi	13·19	2·56	46·5
Shakti (opaque-2)	11·69	3·98	72·4

* Based on lysine as the first limiting amino acid and 5·5 glysine/16 g N in 1973 reference amino acid pattern.

TABLE II

Mean true digestibility, biological value and other properties of proteins of maize varieties

Protein value expressed as a percentage	Casein	Maize varieties			
		Vijay	Ganga-5	Basi	Shakti
True digestibility	100·2±0·5	92·1±0·6	92·8±0·9	93·7±0·9	95·2±0·7
Biological value	84·1±4·4	56·4±2·9	59·9±0·8	56·0±0·9	76·0±1·3
Net protein utilization	84·2	52·2	55·6	52·6	72·3
Utilizable nitrogen	11·42	0·92	1·05	1·11	1·35
N in % of dry matter	13·57	1·76	1·89	2·11	1·87

Nitrogen Balance.—The results of N-balance study are presented in Table II. The casein supplemented with 1% methionine was used as a check. It is evident that the true digestibility (TD) of all the maize varieties under test was more than 90%. The biological value (BV) of the normal maize varieties ranged from 56–60%. The BV of opaque-2 maize was about 35% higher when compared to Basi and Vijay and 27% better than Ganga-5. The net protein utilization (NPU), a derived factor (TD × BV/100) which indicates actual retention by body, was substantially higher for Shakti opaque-2. However, the BV and NPU of opaque-2 maize were found to be 90% and 86% respectively of casein. The utilizable nitrogen (UN = NPU × nitrogen %/100), was found to be the highest for opaque-2 maize.

Among normal maize varieties, Basi had higher UN value (1·11) compared to Ganga-5 (1·05) and Vijay (1·92). Bressani[11] in his studies in Guatemala with 2–6 year old children found the protein efficiency of opaque-2 maize to be 90% of that of skim milk. Similarly Merts[12] in his tests with rats observed that opaque-2 maize had a PER value to be 62%–110% superior to that of ordinary maize.

Feed Efficiency Ratio.—Since the experimental data at hand clearly indicated nutritional superiority of opaque-2 maize the FER of opaque-2 maize was compared with two of the popularly used baby foods, Amulspray and Balamul and a pre-school children supplementary food Balahar. These materials were fed to rats at their inherent protein levels with a view to determining the suitability of opaque-2 maize as a supplementary food for infants and pre-school children. Protein contents of the diets, the total weight gain by rats and FER values are presented in Table III. Opaque-2 maize used in the study contained 11·8% protein.

TABLE III

Mean feed efficiency ratio (FER) of opaque-2 maize diet, Amulspray, Balamul and Balahar in a 28 day rat feeding experiment

Diet source	Protein in % of dry matter (diet)	Average weight gain (g)	FER
Shakti* (opaque-2)	11·18	54·4±4·8	4·01±0·21
Amulspray	24·08	88·5±7·7	2·14±0·05
Balamul	23·46	96·2±8·3	2·38±0·17
Balahar	25·69	43·3±4·6	4·24±0·16

* Opaque–2 maize was fortified with 1% vitamin and 4% mineral mixture.

It is seen that the FER value for opaque-2 maize diet is of the same order as Balahar. This indicates that opaque-2 maize, after supplementation with minerals and vitamins, could be a suitable material for pre-school children. The FER values for Balamul and Amulspray were found to be 60% lower than for opaque-2 diet. The total weight gain with Balamul and Amulspray was much higher compared to Balahar and opaque-2 maize diet. Such results are not entirely unexpected since the total protein content of Amulspray as well as of Balamul is considerably much higher than in opaque-2 maize. Moreover, the protein of the baby food is of animal origin. In terms of economic and the possibility of rapid production of opaque-2 maize, the latter hold considerable promise.

ACKNOWLEDGEMENTS

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1. Mertz, E. T., Bates, L. S. and Nelson, O. E., *Science*, 1964, 145, 279.
2. Munck, L., Karlson, K. E. and Hagberg, A., *Proc. Barley Genet. Sym.*, Washington State University Press, Washington, 1969, p. 544.
3. Singh, R. and Axtell, J. D., *Crop Science*, 1973, 13, 535.
4. Jimenez, J. R., *Proc. High Lysine Corn Conf.* (E. T. Mertz and O. E. Nelson, Ed.), Corn Refineries Association, Inc., Washington, 1966, p. 74.
5. Mehta, S. L., Srivastava, K. N., Mali, P. C. and Naik, M. S., *Phytochemistry*, 1972, 11, 937.
6. Misra, P. S., Jambunathan, R., Mertz, E. T., Glover, D. V., Barbosa, H. M. and McWhirter, K. S., *Science*, 1972, 176, 1425.
7. Gopalan, C., Balavady, B. and Krishnamurthi, B., *Lancet*, 1969, ii, 956.
8. A.O.A.C., *Official Methods of Analysis of the Association of Official Agricultural Chemists*, 10th Ed., 1965, p. 744.
9. Mitchell, H. H. and Carman, G. G., *J. Biol. Chem.*, 1926, 68, 183.
10. Eggum, B. O., "Experimental technique for rats," In : *A Study of Certain Factors Influencing Protein Utilization in Rats and Pigs* (I Kommission hos Landhusholdningsselskabets forlag, Rolighedsvej 26, 1958, Copenhagen, V), 1973, p. 17.
11. Bressani, R., *Proc. High Lysine Corn Conf.*, Corn Refineries Association, Inc,, Washington, 1966, p. 34.
12. Mertz, E. T., *Ibid.*, Corn Refineries Association, Inc., Washington, 1966, p. 12.

PROBLEMS ON HOST SPECIFICITY IN INSECTS

T. N. ANANTHAKRISHNAN
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A THREE-DAY symposium was convened by Dr. T. N. Ananthakrishnan, Director, Entomology Research Unit, Loyola College, Madras, from 24th–26th, January 1976, wherein participants from varied disciplines such as Agricultural Entomology, Botany, Zoology, Medical and Veterinary parasitology presented papers.

In introducing the subject, Dr. T. N. Ananthakrishnan stressed the need for an understanding not only of the host range spectrum of pest species, but also their fecundity, comparative growth rate and population dynamics on the varied hosts in order to correctly assess their role as pests. Among several instances he cited from his own work on thrips, he mentioned about *Haplothrips ganglbaueri* known in recent years to seriously infest paddy inflorescence in many areas and how a direct correlation existed between the infestation of this species in the weed *Echinochloa crusgalli* and *Oryza sativa.*

In his key note address on 'Insect nutrition and host plant selectios,' Dr. N. C. Pant. Head of the Division of Entomology, I.A.R.I., New Delhi, emphasised the need for basic research and exhorted research workers to investigate aspects of nutrition of the insects of crop plants in depth, to elucidate the factors in the host plants that influence their choice of the pests.

In the three days sessions, about 32 papers were presented and three special lectures delivered. The technical papers covered a wide range of insect and acarine pests and parasites of importance in agriculture, medical, veterinary Entomology and included a discussion of the stimuli in the host plants or animals available to the insects for host selection and the response in them to exercise the specific choice. The groups so dealt with were the aphids, aleyrodids, tingids, membracids, pyrrhocorids, thrips, acridids, midges, anthomyids and mites of cotton, sorghum, millets, rice and other economic plants ; and mosquitoes, fleas, cimicids, mallophagans, ticks and mites ectoparasitic on man, birds and other animals. While the contributions discussed host preferences such as monophagy, oligophagy and polyphagy in terms of morphological, physical, chemical and biochemical factors in the plants or animals as determinants of such differential behaviour, stress was repeatedly laid on the need for a sound taxonomic basis in host specificity analysis and delineation. In a few papers which dealt with insect vectors of pathogens such as viruses and parasitic helminths, the vectorial specificity and potentials were examined in detail and attributed to physiological criteria within the insects and to ecological conditions bringing the vector and host together. Diverse aspects of host specificity relating to such cecidozoa as thrips, aphids, coccids and midges in relation to gall production, as well as attempts at a biochemical interpretation of gall formation were also discussed.

Among the highlights of the symposium may be mentioned the three special lectures on 'Biological control of pests' by Dr. T. Sankaran, 'Factors influencing vector status and potential in the transmission of Bancroftian Filariasis by Prof. M. Anantaraman and on 'Some genetic aspects of insect specificity' by Prof. M. Dharmarajan. The advantages of utilising biological agents against injurious pests over other known methods were discussed through numerous instances of success, both in the past and in the present. Current knowledge on the existence of three forms of *Culex pipiens* complex and their relation to periodic and subperiodic, and urban and rural strains of *Wuchereria bancrofti* in different parts of the world and how the influence of filarial strains on the infectiousness and infectivity of mosquito vectors could be studied, were discussed. In the third or valedictory lecture it was elucidated, that host specificity like insecticide resistance, is a pre-adaptive genetic trait and that an understanding of the mechanisms involved in it could generate control or eradication procedures or techniques. Drawing upon several examples from human genetics, such as polymorphism and specificity to parasitic infections, it was explained that gene mutations leading to changes in the protein molecules in the host, could confer an immunity to a particular parasite or invader.

The help received from the University Grants Commission and from the Principal, Loyala College, in the conduct of the symposium is gratefully acknowledged.

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LETTERS TO THE EDITOR

A POTENT TECHNIQUE OF MUTAGENESIS

IONISING radiation such as electrons and protons (from interactions of X-rays, γ-rays and neutrons) and certain chemicals are used extensively as mutagenic agents. We report here the results of an exploratory experiment in which high energy heavy ions were used as mutagens.

Thirty six eggs (thickness $\sim$ 15 microns) of a free living nematode *Caenorhabditis elegans* (Bristol Strain) were exposed to radiation from Californium-252 for five minutes. During this time, about 10 fission fragments, 300 alpha particles and a comparable flux of X-rays, γ-rays and neutrons passed through each egg. The energy deposited in each egg is about 100 MeV per fission fragment, 5 MeV per alpha particle and negligible in case of X-rays, γ-rays and neutrons. Fission fragments are expected to cause particularly severe damage along and around their path.

TABLE I

Phenotypes of the progenies of individual worms hatched from eggs irradiated by Californium-252 radiation

Plate No. \ Phenotypes	Dumpy	Long	Unco-ordinated	Blistered	Fluorescent	Others	Mutant/Normal
1	+		+		+		$\sim$ 20%
2					+		$\sim$ 25%
4					+		$\sim$ 20%
6			+		+		$\sim$ 30%
8					+		$\sim$ 10%
9					+		$\sim$ 20%
10		+					$\sim$ 25%
11				+			$\sim$ 15%
14					+		$\sim$ 20%
16					+		$\sim$ 30%
17						+	100%

Nos. 3, 5, 7, 12 and 15 sterile; No. 13 killed.

17 larvae hatched out of the 36 irradiated eggs. These were allowed to grow on individual plates, and observed for the appearance of mutants in their progeny. Five of the worms turned out to be sterile, all the remaining lines yielded mutant progeny as shown in Table I. The worm in plate No. 13 was killed during transfer. The larger number of the mutants picked are fluorescent mutants[1]; others include most of the known mutant types, *i.e.*, dumpy, long, uncoordinated, plistered, etc.[2]. Two of the lines gave mutants with multiple defects. No. 1 is dumpy, fluorescent and uncoordinated and No. 6 is uncoordinated fluorescent. The mutants have bred true in subsequent generations.

Further experiments on the mutagenic action of Californium-252 are in progress. We are grateful to Prof. O. Siddiqi, Dr. P. Babu, Mr. Satpal Singh and Mr. N. K. Singh, for useful discussions.

Tata Institute of Fundamental Research,
Bombay 400 005, *April* 1, 1976.

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1. Babu, P., "Biochemical Genetics of *Caenorhabditis elegans*," *Mol. Gen. Genet.*, 1974, 135, 39.
2. Brenner, S., "The Genetics of *Caenorhabditis elegans*," *Genetics*, 1974, 77, 71.

TEMPERATURE VARIATION OF ULTRASONIC ABSORPTION IN SOME RING COMPOUNDS

WHILE carrying out studies on the effects of relaxation on ultrasonic absorption, measurements have been made on the temperature variation of ultrasonic absorption in some ring compounds. The variation of absorption with temperature in the liquids Piperidine, α-Pipecoline and Cyclo-Octane are presented here.

Measurements of the coefficient of absorption were made at a frequency of 15 MHz and in the temperature range of 10–90° C, by using the conventional Pulse–Technique[1,2]. The liquids used were of BDH make and of AnalaR quality. The sending and receiving transducers were thickness resonant at $\approx$ 5 MHz, and were mounted on two fused quartz rods (which acted as delay lines as well as temperature isolators), separated by variable acoustic path in the liquid. The accuracy was tested from time to time by measuring those liquids whose absorption is known[3]. An absolute uncertainty of $\pm$ 5 p.c. is estimated for the measurement of absorption, ± 0·5 p.c. for frequency and ± 0·1° C for temperature.

The absorption variation with temperature is shown in Fig. 1. In the case of the liquids Piperidine and α-Pipecoline, the absorption is almost constant in the range 10°–50° C and starts rising in the range 50°–90° C, the temperature coefficient of increase being more in the case of α-pipecoline. In the case of cyclo-octane, the absorption decreases continuously in the range 10°–50° C and remains more or less constant afterwards. The anomalous variation of absorption in these liquids may be due to the possible relaxation that occurs in these ring compounds due to the disturbance of the equilibrium between structural isomeric forms having different energies which they may assume due to the flexibility of the ring.

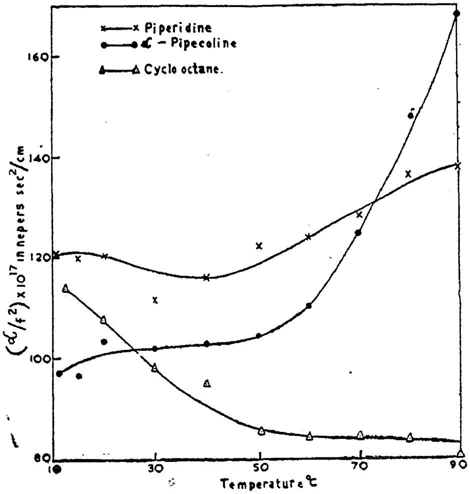

FIG. 1. Temperature variation of ultrasonic absorption in piperidine, α-pipecoline and cyclo-octane at a frequency of 15 MHz.

Thanks are due to Dr. M. G. S. Rao and Prof. B. R. Rao, for encouragement during the course of this work.

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Visakhapatnam-3, *August* 18, 1975.

---

1. Andreae, J. H., Bass, R., Heasell, E. L. and Lamb, J., *Acustica*, 1958, **8**, 131.
2. Pinkerton, J. M. M., *Proc. Phys. Soc.*, London, 1949, **62 B**, 286.
3. Heasell, E. L. and Lamb, J., *Ibid.*, London, 1956, **69**, 869.

COPOLYMERIZATION OF ACRYLIC AND METHACRYLIC ACIDS WITH N-VINYL PYRROLIDONE IN AQUEOUS SOLUTION

ACRYLIC acid (AA) and methacrylic acid (MA) polymerize in aqueous solutions, with a strong dependence of polymerization rate on the degree of ionization and hence on the pH of the medium[1,2]. This is due to the propagation rate constant (k_p) being sensitive to electrostatic interactions between the reaction species during ionization of the monomer and the growing polymer, the termination constant (k_t) being unaffected[1,2]. This dependence on pH will be reflected in the monomer reactivity ratios if these monomers are copolymerized with a nonionic water soluble monomer like N-vinyl pyrrolidone (NVP). The r_1 (of AA or MA) and r_2 (NVP) values were determined by copolymerization in aqueous solution at 30° C in the pH range 2–10 with a view to confirm the above facts and to clarify the conflicting data of r_1 and r_2 reported in the literature in different organic solvents.

Experimental

The copolymers obtained at differest monomer feeds (nearly 1 M aqueous solution) after polymerization by $K_2S_2O_8$–Na_2SO_3 initiator system at each pH to high conversion (5–70%) were isolated quantitatively by concentration of the syrup at room temperature in the presence of excess of hydroquinone and reprecipitation several times by acetone and drying. The copolymers were analyzed for nitrogen by Kjeldahl's semimicro nitrogen estimation procedure[3] and the results were treated by the Mayo–Lewis integrated copolymer equation[4] with a modified computer procedure[5].

Results and Discussion

The reactivity ratios (r_1) for AA and MA are shown as a function of pH in Fig. 1. The r_1 for

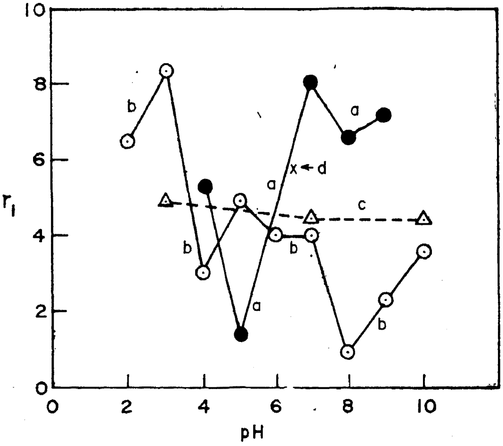

FIG. 1

AA (curve a) shows a minimum of 1 : 3 at pH = 5 at which 86% of the acids is ionized (pKa for AA = 4·2) and then varies between 6·6 and 8 in the pH region 7–9. Addition of 1 M NaCl at pH 6·5 drops r_1 to 5·8 (curve *d*), closer to the r_1 value of predominantly unionized acid at pH = 4. Although the r_1 values at the alkaline pH are expected to be much lower than the value at pH = 4 because of repulsive interactions, they show higher values, which may be partly understood in terms of the homopolymerization rate of acrylic acid in the alkaline medium[2] which is almost comparable to that in the acid medium. The higher values may also be due to the fact that acrylic acid binds 50–65% of the sodium ions[6] in the pH range 6·5–7·5.

The r_1 values of MA show two minima (curve *b*), one at $r_1 = 2·9$ (pH = 4) and $r_1 = 0·92$ (pH = 8) which are obvious from the pKa values[1] of the monomer (pKa = 4·32) and the polymer (pKa = 7·0), the electrostatic repulsion between the reacting species being higher at pH > pKa value of the species. The r_1 increases at higher pH. Addition of a strong electrolyte, 1 M NaCl, reduces the charge interactions, bringing $r_1 \approx$ 4–5 (curve *c*) independent of the pH. This behaviour is also similar to the trend of homopolymerization rate of methacrylic acid[1,2].

Homopolymerization in both systems, is precluded in spite of r_1 values being high, because, as is well known in any copolymerization, the reaction is stopped *before both the monomers* are fully consumed. One has long sequences of the monomer whose $r_1 > 1$.

The high value of r_1 for MA at low pH is obvious because the reacting species are unionized.

The r_2 values obtained for NVP, for both the systems are nearly equal to zero, similar to the values reported in organic solvents[7] although the r_1 for AA in water are considerably higher than those obtained in organic solvents[7].

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---

1. Kabanov, V. A., Topchiev, D. A. and Karaputtadze, T. M., *J. Poly. Sci. Symposium*, 1973, No. 42, 173.
2. —, and —, *Polym. Sci.* (*USSR*), 1971, 13, 1486.
3. Jean, P. Dixon, *Modern Methods in Organic Microanalysis*, D. van Nostrand, Co. Ltd., London, 1968, p. 60.
4. Mayo, F. R. and Lewis, F. M., *J. Am. Chem. Soc.*, 1944, 66, 1594.
5. Joshi, R. M., *J. Macromol. Sci. Chem.*, 1973, 7 (6), 1231.
6. Ferry, G. V. and Gill, S. I., *J. Phys. Chem.*, 1962, 66, 999.
7. Chapiro, A. and LeDoan Trung, *Eur. Polym. J.*, 1974, p. 1103.

CHEMICAL EXAMINATION OF TIMBERS OF MELIA SPECIES

THE family of Melia is considered to be very important in the indigenous system of medicine[1]. The heartwood of the less common Melia species namely *Aglaia andamanica*, *Amoora wallachi* and *Carapa obovata* has been chosen for chemical investigation. All the three species have been obtained from Andaman Islands.

The heartwood of *Aglaia andamanica* was extracted successively with hexane, benzene, diethyl ether, ethyl acetate, acetone and ethanol. The hexane extract gave positive Liebermann-Burchard test for sterol and showed two spots on TLC plate (silica gel, benzene: ethyl acetate 4 : 1). The residue from the hexane extract was fractionally crystallised from methanol to give two crystalline solids. The less soluble compound was obtained as shining needles, m.p. 138°; $(\alpha)_D^{22°}$: − 35° in $CHCl_3$. Its acetate ($NaOAc/Ac_2O$), m.p. 134°; $(\alpha)_D^{22°}$: − 38·5° in $CHCl_3$. Its identity as β–sitosterol was established by comparison with authentic sample.

The more soluble fraction crystallised from ethanol as rhombic crystals, m.p. 217–218°; $(\alpha)_D^{28°}$: −43° in $CHCl_3$; (Found C 69·48, H 7·20 $C_{28}H_{34}O_7$ requires C 69·7, H 7·05%); λ_{max}^{MeOH} 215 (4·12) and 335 (1·8); ν_{max}^{Nujol} 3500, 1740, 1668, 1500 and 875 cm^{-1}; NMR in $CDCl_3$ (δ values) 1·07 (6H), 1·16 (3H), 1·22 (3H), 1·25 (3H) (methyl groups), 2·10 (s, Ac), 7·45 (d, 1H), 6·40 (t, 1H) (α, β protons of furan ring), other protons at 3·53 (s), 4·58 (m), 5·63 (s), 5·86 (d) and 7·15 (d). With 2 : 4-dinitrophenyl hydrazine reagent, the compound formed a derivative which crystallised from methanol as red needles melting at 254–255° (Found C 61·35, H 5 55, N 8·1 $C_{33}H_{38}O_{10}N_4$ requires C 61·6, H 5·80 and N 8·4%). The compound agreed in description in all respects with gedunin[2,3]. When admixed with an authentic sample of gedunin, its mixed melting point* remained undepressed.

The benzene, diethyl ether, ethyl acetate, acetone and ethanol extracts when examined individually on TLC plate using different solvent systems (benzene: hexane 1 : 1 benzene, benzene: diethyl ether 1 : 1, benzene : ethyl acetate 4 : 1, chloroform : methanol 95 : 5 and 9 : 1) did not show any spots but appeared as continuous streaks. The individual extracts were separated into sodium bicarbonate (2%) soluble, sodium carbonate (2%) soluble and sodium hydroxide (2%) soluble fractions. This fractionation also did not help to induce the material to crystallise although the ethyl acetate, acetone and ethanol extracts gave tests for phenolic compounds with ferric chloride.

The hexane extract of *Amoora wallachi* answered Liebermann-Burchard test for the presence of sitosterol. The residue from the hexane extract crystallised from methanol as needles, m.p. 138°. $(\alpha)_D^{22°}$; $-35°$ ($CHCCl_3$). The benzene, ether, ethyl acetate, acetone and ethanol extracts behaved in a similar way as in the case of the wood of *Aglaia andamanica*.

The heartwood of *Carapa obovata* was extracted and examined in a similar way. Its hexane extract gave β-sitosterol and gedunin.

Our thanks are due to Prof. T. G. Halsall, Dyson Perrins Laboratories, Oxford, U.K., for the identification of gedunin. Two of us (K.S. and J.M.R.) are thankful to C.S.I.R. for research fellowships.

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* M.M.P. determination was kindly carried out by Prof. T. G. Halsall.

1. Kirtikar, K. R. and Basu, B. D., *Indian Medicinal Plants*, Vol. I, p. 533.
2. Akisanya, A., Bevan, C. W. L., Hirst, J., Halsall, T. G. and Taylor, D. A. H., *J. Chem. Soc.*, 1960, p. 3827.
3. Powell, J. W., *J. Chem. Soc., C Org.*, 1966, **20**, 1794.

2-HYDROXY-5-METHYLACETOPHENONE SEMICARBAZONE AND THIOSEMICARBAZONE AS METAL INDICATORS FOR THE COMPLEXOMETRIC TITRATION OF IRON (III) WITH EDTA

THOUGH Variamine Blue B is the recommended indicator for the volumetric titration of iron (III) with EDTA at pH 2–3[1], it has the disadvantage that (*i*) even mild oxidizing agents like nitrite interfere, and (*ii*) a part of iron should be in bivalent state and this does not react with EDTA. The present note describes the use of 2-hydroxyacetophenone semicarbazone and thiosemicarbazone as metal indicators for the complexometric titration of iron (III) with EDTA. The optimum conditions worked out are the following:

Adjust the pH of iron (III) solution to 2 with ammonia; and 10 ml of 1 *M* hydrochloric acid-sodium acetate buffer of pH 2, and 5 to 10 drops of 0·5% w/v ethanolic solution of the indicator (*i*) 2-hydroxy-5-methylacetophenone semicarbazone, or (*ii*) 2-hydroxy-5-methylacetophenone thiosemicarbazone. Dilute to 100 ml with distilled water and titrate against the standard solution of 0·005 to 0·050 *M* EDTA till a sharp colour change from green to colourless in the case of (*i*) and from bluish green to colourless in the case of (*ii*) is observed.

With higher amounts of iron (III), the end point is indicated by the disappearance of the last traces of greenish colour leaving behind a clear yellow solution.

The relative standard deviation for the titration has been found to be 0·3–0·5% for 1–70 mg iron (III).

With 100 mg and above of iron (III), the standard deviation was found to be nearly 1%.

Interferences

The following ions do not cause any interference even when present in quantities upto 100 times (w/w) that of iron (III): bromide, sulphate, chloride, tartrate, citrate, urea, calcium, magnesium, barium, strontium, beryllium, zinc, cadmium, mercury (II), manganese-(II), uranyl (VI) and nitrite.

The following ions can be tolerated in amounts given in parentheses, together with masking agents if any: 4·90 iron (III) (4·9 mg): aluminum (III) (200 mg. tartrate), antimony (III) (50 mg, tartrate), bismuth (III) (50 mg, tartrate or urea with end point as the disappearance of green colour), chromium (III) (20 mg with end point as the disappearence of green colour), cobalt (II) (30 mg, tartrate), fluoride (50 mg), molybdate (VI) (100 mg, tartrate or citrate), oxalate (20 mg with end point as the disappearance of green colour), phosphate (200 mg), thiosulphate (200 mg), titanium (IV) (20 mg, tartrate) and zirconium (IV) (100 mg, tartrate).

The following ions interfere seriously: cyanide, sulphocyanide, copper (II), nickel (II) and vanadium (IV, V).

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September 26, 1975.

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1. Vogel, A. I., *A Text Book of Quantitative Inorganic Analysis*, Longmans–Green, London, 1962, (3rd. Edn.), p. 444.

FLAVANOIDS OF *ACACIA SUMA* HEARTWOOD QUERCETIN AND HYPERIN 3′, 7-DIMETHYL ETHER

THE fine shavings of the heartwood of *Acacia suma* were extracted with boiling ethanol. The concentrated ethanolic extract was fractionated into ether and ethyl acetate soluble fractions. The ether extract gave a good yield of a flavanoid compound which was characterised as quercetin by comparison with authentic sample. The concentrated ethyl acetate extract was subjected to fractional precipitation with petroleum ether and after removal of the resinuous impurities, a chromatographically homogeneous flavanoid glycoside, could be isolated.

Hyperin 3′, 7-dimethyl ether

It was isolated as a pale yellow solid, m.p. 190–192°. It gave positive Molisch test suggesting the presence of a carbohydrate moiety in the molecule. It neither reduced Fehling solution nor gave any positive colour with aniline hydrogen phthalate, which showed that the reducing group of the sugar is involved in the glycosidic linkage. Acid hydrolysis of the glycoside gave a water insoluble aglycone and a reducing sugar which was identified as D (+) galactose by mixed paper chromatography, R_f value of its benzyl amine derivative and mixed m.p. of its osazone.

The pale brown aglycone $C_{17}H_{14}O_7$; m.p. 214°, was found to be flavanoid in nature by its characteristic colour reactions and positive colour reaction with zirconium oxychloride in presence of citric acid. The aglycone was analysed for two methoxyl groups (Zeisel's method) as supported by the IR peak at 2850 cm^{-1}. With acetic anhydride and pyridine the aglycone formed a triacetate thereby the presence of three hydroxyl groups was indicated. The relative positions of the methoxyl and hydroxyl groups was determined by the colour reactions, spectral shifts[2] and degradative studies. The tetramethyl ether of the glycoside on oxidation with neutral potassium permanganate gave veratric acid as one of the oxidation products, which suggested that the side phenyl is a 3 : 4 substituted nucleus. The absence of a free phloroglucinol unit in the aglycone was indicated by the absence of a positive colour reaction with vanillin-hydrochloric acid reagent. However, its positive test with ethanolic boric acid in the presence of citric acid showed the presence of a free hydroxyl group at 5 position. No bathochromic shift on addition of sodium acetate indicated that probably 7 hydroxyl group is substituted. Therefore, one of the methoxyl groups eventually was located at position 7. A bathochromic shift was observed on addition of ethanolic aluminium chloride which showed the presence of free hydroxyls at position 3 and 5. On addition of sodium ethylate (0·002 M) long wavelength band at 374 nm disappeared which indicated the presence[3] of free hydroxyls at 3 and 4′. Therefore, 5, 3, and 4′ positions carry free hydroxyls and conclusively one of the two methoxyl groups must be present in position 3′ of the side phenyl.

The position of glycosidic linkage was ascertained by comparing the colour reactions of the aglycone with those of the glycoside. The glycoside failed to give reaction with zirconium oxychloride in presence of citric acid[4] whereas aglycone responded to it, indicating thereby that 3-hydroxyl of the aglycone is involved in glycosidic linkage.

The glycoside could be hydrolysed to give free sugar and aglycone with emulsin isolated from almonds. Hence it is concluded that the substance is a β-glycoside. The ring structure of the galactose moiety was found to be pyranose, as the glycoside methyl ether with all the free phenolic hydroxyls methylated on periodate oxidation consumed two molecules of periodate with liberation of one molecule of formic acid.

Thus, it has been found to be 5, 4′-dihydroxy-7, 3′-dimethoxy flavone 3–O–β–D–galactopyranoside or 3′, 7-dimethyl flavone ether of hyperin.

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---

1. Briggs, L. H., Colebrook, L. D., Fales, H. M. Wildman, W. C., *Anal. Chem.*, 1954, **29**, 904.
2. Jurd, L. and Geissman, T. A., *J. Org. Chem.*, 1956, **21**, 1395.
3. —, In : *Chemistry of Flavonoid Compounds*, Edited by T. A. Geissman, Pergamon Press, Oxford, 1962, p. 107.
4. Hörhammer, L. and Hänsel, R. M., *Arch. Pharm.*, 1955, **288**, 315.

SOME METAL BISTHIOSALICYLATO MERCURATES

THE preparation and studies on mercuric thiosalicylate has already been described[1]. In the present communication, preparation and analysis of cerium, chromium and thallium bisthiosalicylato mercurates are reported. The preparation[2] and other studies[3] on disodium bisthiosalicylato mercurate have also been discussed.

Chromium solution was prepared from $CrCl_3 \cdot 6\,H_2O$ which was a 'Baker Analyzed' reagent. Thiosalicylic acid was from Evans Chemetics (U.S.A.). All other chemicals and reagents were of B.D.H. (India) Analar quality. Cerium and chromium bisthiosalicylato mercurates were obtained on mixing the equivalent quantities of 0·01 M solutions of disodium bisthiosalicylato mercurate and the metal ions. Thallium mercurate precipitated only after adding tenfold excess of the thallium solution. Cerium gave a buff coloured precipitate and a light-grey chromium mercurate was obtained, measurement of absorption, ± 0·5 p.c. for frequency and ± 0·1° C for temperature.

The absorption variation with temperature is shown in Fig. 1. In the case of the liquids Piperidine and α-Pipecoline, the absorption is almost constant in the range 10°–50° C and starts rising in the range 50°–90° C, the temperature coefficient of increase being more in the case of α-pipecoline. In the case of cyclo-octane, the absorption decreases continuously in the range 10°–50° C and remains more or less constant afterwards. The anomalous variation of absorption in these liquids may be due to the possible relaxation that occurs in these ring compounds due to the disturbance of the equilibrium between structural isomeric forms having different energies which they may assume due to the flexibility of the ring.

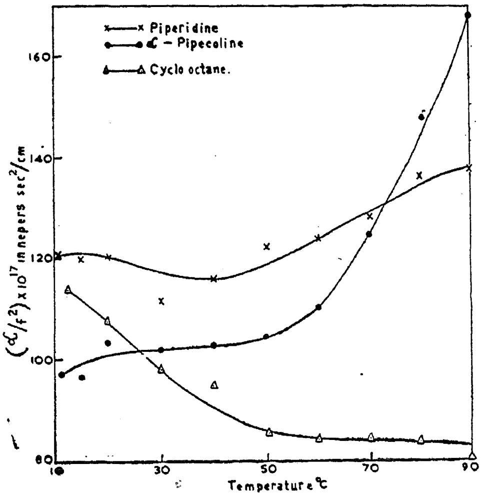

FIG. 1. Temperature variation of ultrasonic absorption in piperidine, α-pipecoline and cyclo-octane at a frequency of 15 MHz.

Thanks are due to Dr. M. G. S. Rao and Prof. B. R. Rao, for encouragement during the course of this work.

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---

1. Andreae, J. H., Bass, R., Heasell, E. L. and Lamb, J., *Acustica*, 1958, **8**, 131.
2. Pinkerton, J. M. M., *Proc. Phys. Soc.*, London, 1949, 62 **B**, 286.
3. Heasell, E. L. and Lamb, J., *Ibid.*, London, 1956, **69**, 869.

COPOLYMERIZATION OF ACRYLIC AND METHACRYLIC ACIDS WITH N-VINYL PYRROLIDONE IN AQUEOUS SOLUTION

ACRYLIC acid (AA) and methacrylic acid (MA) polymerize in aqueous solutions, with a strong dependence of polymerization rate on the degree of ionization and hence on the pH of the medium[1,2]. This is due to the propagation rate constant (k_p) being sensitive to electrostatic interactions between the reaction species during ionization of the monomer and the growing polymer, the termination constant (k_t) being unaffected[1,2]. This dependence on pH will be reflected in the monomer reactivity ratios if these monomers are copolymerized with a nonionic water soluble monomer like N-vinyl pyrrolidone (NVP). The r_1 (of AA or MA) and r_2 (NVP) values were determined by copolymerization in aqueous solution at 30° C in the pH range 2–10 with a view to confirm the above facts and to clarify the conflicting data of r_1 and r_2 reported in the literature in different organic solvents.

Experimental

The copolymers obtained at differest monomer feeds (nearly 1 M aqueous solution) after polymerization by $K_2S_2O_8$–Na_2SO_3 initiator system at each pH to high conversion (5–70%) were isolated quantitatively by concentration of the syrup at room temperature in the presence of excess of hydroquinone and reprecipitation several times by acetone and drying. The copolymers were analyzed for nitrogen by Kjeldahl's semimicro nitrogen estimation procedure[3] and the results were treated by the Mayo–Lewis integrated copolymer equation[4] with a modified computer procedure[5].

Results and Discussion

The reactivity ratios (r_1) for AA and MA are shown as a function of pH in Fig. 1. The r_1 for

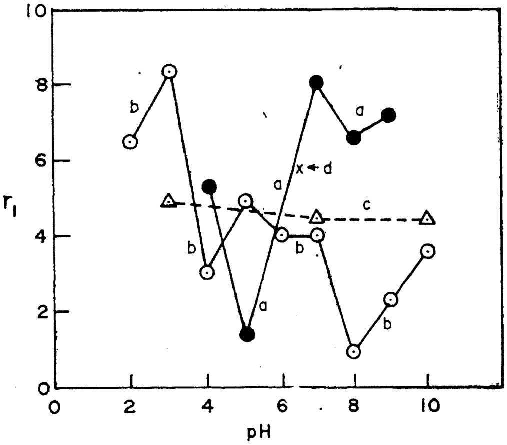

FIG. 1

Thallium gave a dirty white precipitate. The reaction mixtures were digested on a water bath and kept overnight. The precipitates were filtered, washed with water and dried in oven at 100° C for two hours. The dried mercurates were analysed for mercury and the sulphur contents. The experimental values of sulphur and mercury corresponded well with the theoretical values.

The mercurates were characterised on the basis of their Nujol mull infrared spectra, recorded in 4600–700 cm^{-1} region on a Specord-71, Spectrophotometer. Bands corresponding to —SH frequencies, observed in the case of thiosalicylic acid, disappear completely in the spectra of the mercurates. But the bands corresponding to $C = O$ stretching vibrations shift to lower frequency and are observed to split. $C - S$ stretching vibrational bands also shift to a lower frequency. The disappearance of —SH frequency band and the lowering of $C - S$ stretching frequency indicate the coordination through sulphur atom. The coordination through carboxyl oxygen and carboxylic group is indicated by lowering of $C = O$ stretching frequencies. The splitting of the bands may be due to coupling of some vibrations.

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---

1. Dubey, K. P. and Koul, M. K., *Rev. Chem. Miner.*, 1971, 8, 819.
2. Koul, M. K. and Dubey, K. P., *J. Inorg. Nucl. Chem.*, 1973, 35, 2567.
3. — and —, *Ibid.*, 1973, 35, 2571.

NOTE ON THE DISCOVERY OF *CONOPHYTON* AND THE RECORD OF ONCOLITES FROM THE KUMAON HIMALAYA

THE present note records the discovery of the stromatolite *Conophyton* from the Kumaon Himalaya as a whole, and oncolites from the Calc Zone of Tejam. The typical cone-in-cone type of algal structure (Plate Figs. 1 to 4), as recorded here, belongs to the form *Conophyton cylindricus* Maslov, so typical of the Lower Riphean. Oncolites have so far been mainly reported from Siberia and Japan only. Report of oncolites from the Precambrian terrains of India is very scarce to almost lacking.

The above-mentioned forms have been recorded from the thick calc-argillaceous Kapkot formation (Misra and Bhattacharya, 1972) of the Calc Zone of Tejam, around Kapkot village (29° 56′ 19″ : 79° 54′ 9″) in the northern Almora District. The host rocks in which the forms occurs, are grey dolomitic limestones.

Conophyton cylindricus occurs as conical to sub-cylindrical forms, disposed vertically. They taper towards the top and are typically constricted near the base (Plate Figs. 1, 3). The transverse section is circular to elliptical and the cross-section shows dark and light coloured laminae arranged in nested cones (Plate Figs. 2, 4). They contain a prominent central or axial cone surrounded by concentric, but thinner, conical laminae. Two varieties have been distinguished.

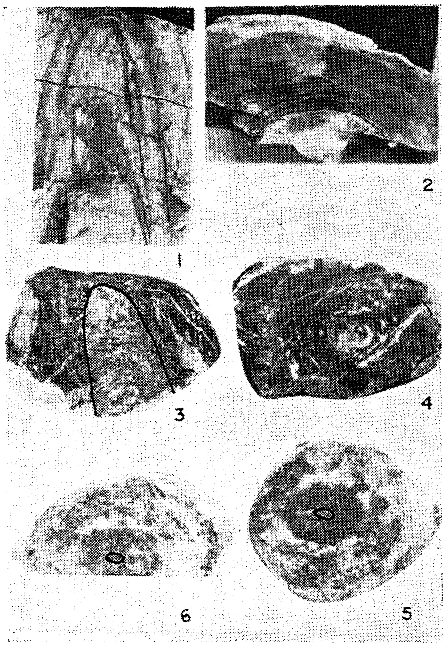

PLATE FIGS. 1–6.

Variety 1 :

Length is about 14 cm. ; diameter at the base is about 12·5 cm. ; axial zone is about 5·5 cm. wide at the base. Apical angle of the cones is 25° to 30°.

Variety 2 :

Length is about 7 cm. ; diameter at the base is about 6 cm. The axial zone is about 2·8 cm. wide at the base. Apical angle of the cones is about 35°.

The major differences between var. 1 and var. 2 are that the var. 2 is (*i*) smaller in size and (*ii*) darker in colour, than var. 1.

The oncolites typically occur as discrete spheroids (Plate Figs. 5, 6). The constituting units are randomly stacked circular (to sub-elliptical) laminae, arranged concentrically around a central nucleus. The longest and the shortest axes of the forms are 4 cm. and 3·5 cm. respectively. These structures are seen on the upper surfaces of the beds, and are not seen in vertical section. They are closely associated with *Conophyton cylindricus* and *Collenia*.

Work on oncolites, though rare, reveals that they are distributed throughout the stratigraphic column (Konishi, 1949; Johnson, 1963; Laporte, 1963).

Conophyton cylindricus has been described from South Africa, Sahara, Siberia, North America and China. In India, they have been reported from the Vindhyans (Fawn Limestone of the Semri Series) (Valdiya, 1969), the Jammu Limestone (Raha and Sastry, 1973) and from the Bijawars of Madhya Pradesh (Krishna Murthy, 1972).

Conophyton has been regarded as an important and reliable index fossil of the Precambrian (Komar *et al.*, 1965; Bertrand–Sarfati, 1970, 1972; Koroljuk, 1960; Raaben, 1969). Particularly, the form *Conophyton cylindricus* is characteristic of Lower Riphean (Krylov, 1960; Keller *et al.*, 1960; Zhuravleva and Komar, 1962). As a particular case, the relatively smaller forms of *C. cylindricus* with 6–8 cm. diameter are taken to be typical of the Lower Riphean within the absolute age limits of 1450–1430 m.y. (Komar *et al.*, 1965).

On the basis of *Conophyton cylindricus*, the Kapkot formation of the Calc Zone of Tejam, can easily be assigned a Lower Riphean age, and now be easily be correlated with the Jammu Limestone, the Semri Series (Vindhyans) and the Bijawars. Further, by analogy, the possible equivalents of the Kapkot formation (Bhattacharya, 1973, p. 37), *viz.*, Thalkedar Limestone of the Calc Zone of Pithoragarh, Upper Shali Limestone of the Simla Himalaya and the Krol Limestone of the Solon–Tons area, may also be assigned a Lower Riphean age.

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Department of Geology, A. R. BHATTACHARYA.
Lucknow University,
Lucknow, *September* 23, 1975.

1. Bertrand–Sarfati, J., *Palaeogeogr., Palaeoclimat., Palaeoecology*, 1970.
2. —, *Musee Royal de L'Afrique Centrale-Tervuren, Belgique-Annales, Serie IN–8°–1 Sciences Geologiques–74*, 1972.
3. Bhattacharya, A. R., *Ph.D. Thesis*, Unpublished, Lucknow University, 1973.
4. Johnson, J. H., *Quat. Colorado School Mines*, 1963, **58** (3).
5. Keller, B. M. *et al.*, *Izvestia Akademii Nauk SSSR*, Ser. Geol., 1960, 2, 21.
6. Komar, V. A. *et al.*, *Dokl. Nauk SSSR*, 1965, 131, 73.
7. Konishi, K., *Jour. Fac. Sci. Univ. Tokyo*, Sec. II, 1959, 7 (4), 441.
8. Koroljuk, I. K., *Rept. 21st Intern. Geol. Congr.*, 1960, **8**, 113.
9. Krishna Murty, M., *Jour. Geol. Soc.*, India, 1972, 13 (2), 181.
10. Krylov, I. N., *Akad. Nauk SSSR, Dokl.*, 1960, Earth Sci. Sec., in trans., 126 (1–6), 512.
11. Laporte, L. F., *Jour Paleont.*, 1963, 37 (3), 643.
12. Misra, R. C. and Bhattacharya, A. R., *Himalayan Geol.*, 1972, 2, 252.
13. Raaben, M. E., *Amer. Jour. Sci.*, 1969, **267**, 1.
14. Raha, P. K. and Sastry, M. V. A., *Himalayan Geol.*, 1973. 3, 135.
15. Valdiya, K. S., *Jour. Geol. Soc.*, India, 1969, 10 (1), 1.
16. Zhuravleva, Z. A. and Komar, V. A., *Dokl. Akademii Nauk SSSR*, 1962, 134, 197.

CHANGES IN AMINO ACIDS AND AMIDES INDUCED BY PAPAYA MOSAIC VIRUS IN PAPAYA PLANTS

PAPAYA mosaic virus (PMV) causing a very serious disease of papaya was reported from this laboratory by Capoor and Varma (1958). PMV-infected papaya plants are stunted and bear mosaic mottled, feformed and reduced leaves. The virus is mechanically transmissible and aphid borne. PMV is transmitted by aphids in stylet borne manner (Capoor and Varma, 1958; Singh, 1972) and also infects many cucurbitaceous plants (Capoor and Varma, 1958). Twenty to twenty-five days old healthy and vigorously growing *Carica papaya* Linn. cv 'Washington' plants were used for extraction and determination of free amino acids and amides. Fifty plants inoculated with PMV by sap inoculation method and fifty uninoculated control were kept under insect proof glass house. After one month of inoculation leaf (four youngest), stem and root samples were collected from both control and diseased papaya plants and amino acids and amides were determined both qualitatively and quantitatively by the method described by Thankappan and Chacke (1970).

As obvious from Table I, the quantity of amino acids and amides was maximum in leaves and minimum in roots. In healthy papaya plants

TABLE I

Amino acids, and amides present in the leaves, stems and roots of healthy and PMV-infected papaya plants

Amino acids and amides	µg/g of tissue					
	Leaf		Stem		Root	
	H	D	H	D	H	D
Aspartic acid	112	163	30	47	19	12
Glutamic acid	146	152	48	55	9	18
Glycine-serine	10	10	7	10	Trace	1
Lysine	1	5	..	2	..	Trace
Aspargine	12	48	7	20	3	2
Threonine	25	37	10	7	15	13
Argenine	12	6	..	..	..	..
Alanine	57	190	71	65	..	..
Glutamine	..	..	15	27	3	9
B-alanine	..	..	5	..	..	..
Homoserine	Trace	Trace	3	6	..	2
Thryptopane	1	Trace	4	2	..	Trace
Methionine	..	6	Trace	Trace	Trace	..
Hystidine	..	3	..	..	—1	2
Tyrosine	..	1	..	2	2	1
Proline	Trace	..	..	1	Trace	Trace
Y-amino butyric acid	2	1	Trace	Trace	1	7
Valine	6	9	..	..	2	1
Phenylalanine	9	11	..	..	3	5
Leucine, isoleucine	Trace	7	8	20	Trace	9
Total amount of amino acids and amides	393	649	208	264	58	82
Total number of amino acids and amides	12	15	11	13	10	13

H = Healthy, D = Diseased.

respective concentration of amino acids and amides per gm. of leaves, stems and roots were 393, 208 and 58 µg and the quantities present in the leaves, stems and roots of PMV-infected papaya plants were 649, 264 and 82 µg per g. of tissue respectively. Thus PMV infection greatly increased the quantity of the amino acids and amides. In general leaves and stems contain large quantities of aspartic acid, glutamic acid, aspargine, threonine, argenine and alanine. The other major amino acids present in leaves were glycine-serine, valine and phenylalanine. Stems also contain considerable amounts of glycine-serine, glutamin, leucine and iso-leucine. Glutamic acid was higher in the roots of the infected than the healthy papaya plants. In addition to aspartic acid, glutamic acid and threonine, roots also contained considerable amounts of aspargine, glutamine and phenylalanine. It was also observed that total number of amino acids and amides present in leaves, stems and roots of PMV-infected papaya plants were much greater than those present in the corresponding part of healthy papaya plants (Table I). Similar results have also been obtained by other workers in other host-virus combinations (Diener, 1960; Narayanswamy and Ramakrishnan, 1960; Welkie *et al.*, 1967; and Thankappan and Chacko, 1970).

Accumulation of amino acids in virus-infected plants may be due either to the increased proteolytic activity or due to the inhibition of normal protein synthesis. For accelerated synthesis of virus protein large quantities of amino acids are required and virus infection increases the amino acids both qualitatively and quantitatively so that synthesis of virus protein may occur without any hindrance.

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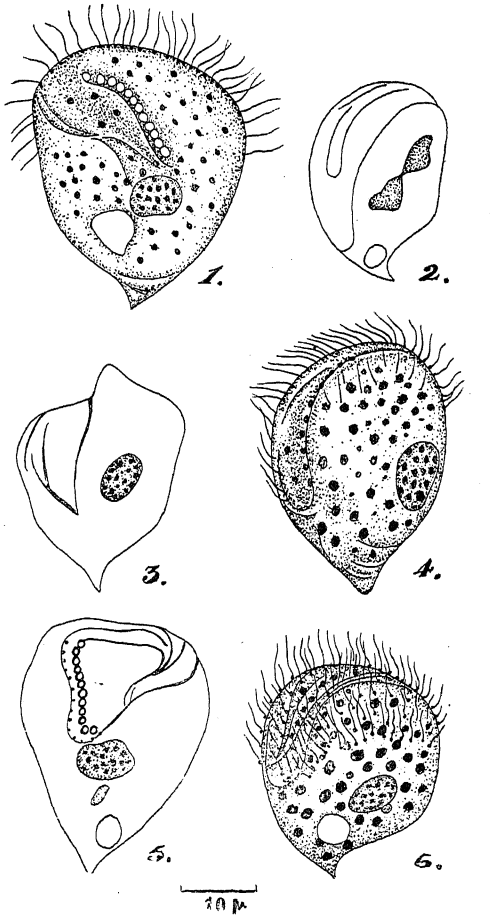
1. Capoor, S. P. and Varma, P. M., *Ind. J. Agric. Sci.*, 1958, 28, 225.
2. Diener, T. O., *Phytopathology*, 1960, 50, 198.
3. Narayanswamy, P. and Ramakrishnan, K., *Proc. Ind. Acad.* 1966, 63 B, 288.
4. Singh, A. B., *Ind. J. Ent.*, 1972, 34, 240.
5. Thankappan, M. and Chacko, C. I., *Ind. J. Pl. Physiol.*, 1970, 13, 99.
6. Welkie, G. W., Yang, S. F. and Miller, G. W., *Phytopathology*, 1967, 57, 469.

A NEW PARASITIC CILIATE *CYATHODINIUM INDIAE* N. SP., FROM THE GUINEAPIG, INDIA

DURING the course of the systematic survey of the parasitic protozoa in the birds and mammals of the Hyderabad region, a very interesting ciliate of the genus *Cyathodinium* was found in the laboratory bred guineapig *Cavia cutleri* which died of some unknown cause. This ciliate was encountered along with some other flagellates. However, the infection was very heavy. Subsequently a few more guineapigs were dissected but the infection was negative. Davine[1] (1875), Nie[2] (1950) and others have extensively studied the parasites of guineapig and have reported a number of flagellates and a few ciliates. However, the present *Cyathodinium* differs totally from the species so far described and this is the first report from India.

Cyathodinium n. sp. was fixed in Schaudinn's fixative and stained in iron haematoxylin. Few specimens were studied under vital stains. It is funnel shaped abruptly ending into a pointed tip which sometimes curved to one side (Figs. 1, 2, 3, 5, 6). It measures $55\,\mu$ in length and $20\,\mu$ in width. Rarely the nipple-like tip is also observed (Fig. 4). The pseudoperistome extends from one side of the anterior part of the parasite to half or more than half of the body length (Figs. 2, 4). The pseudoperistome has two conspicuous lips of which the left one appears to overlap the right (Fig. 3). Endosprits, which are in a definite number of twelve, are situated either on the left lip or on the right lip. The cilia are present on both the lips (Figs. 1, 4–6), but they do not arise from the endosprits. The cytostome is at the terminal end of the pseudoperistome as against the statement of Cunha and Freitas[3] (1940), who regarded this parasite as an Astomatous ciliate. The cilia restricted to the anterior part only and are arranged clearly in three to four rows (Fig. 6). The position of the macronucleus is variable and is at the middle or at the posterior third of the body (Figs. 1 and 4). It is oval in shape with large clear chromatin granules. The micronucleus is spherical and is very close to the macronucleus (Fig. 6). Divisional forms are also noticed. In these forms a deeply stained rod which Cunha and Freitas called as ‘cristalide” is also seen. The two separated macronuclei are connected by this rod (Fig. 2). During division the nuclei are irregular in shape. Cysts are not observed.



FIGS. 1–6 fixed in Schaudinn's fluid and stained with iron haemotoxylin. Fig. 1. Showing endosprits, nucleus and contactile vacuole. Fig. 2. Showing the dividing nucleus. Fig. 3. Showing the cytostomal lips. Fig. 4. Showing the pseudoperistome and cilia. Fig. 5. Showing the funnel-shaped body and endosprits. Fig. 6. Showing the cilia.

Though *Cyathodinium* species are described by Lucas[4] (1932), Cunha[5] (1914) and Nie[2] (1950) from guineapig, there is no record of it from India. The new species comes very close to *C. piriforme* Cunha and *C. cunhai* Nie. Howerver in shape it differs from the latter. The pseudoperistome is conspicuous and very large extending 3/4 of the body length and has deeply stained lips but in known forms they are very short. Endosprits are definite in number whereas the other species have 9–15 endosprits. The cilia is *C. pyriforme* are very long and extend throughout the anterior half of the body and in *C. cunhai* they are arranged in two rows while in new form they are in 3 or 4 rows. In both the known species the round macronucleus is central in position but in the new species it is oval and is at the posterior end of the body. The contractile vacuole is at the posterior tip of the body in new species whereas in the known forms it is at the posterior dorsal region of the body, but not at the tip.

In view of these differences the present species is a new one, so it is designated as *Cyathodinium indiae* n. sp.

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---

1. Davaine, C., *Dict. Encycl. Sci. Med.*, 1875, 9, 115.
2. Dashu, Nie., *J. Morphology*, 1950, 46, 381.
3. Cunha, A. M. DA and De E, Freitas, C. R., *Soc. Biol.*, 1936, 123, 436.
4. Lucas, M. S., *Arch. f. Protist.*, 1932, 77, 64.
5. Cunha, A. M. DA., *Mem. do Inst. Oswaldo Cruz.*, 1914, 6, 212.

ACANTHOSQUILLA TIGRINA (NOBILI) (CRUSTACEA : STOMATOPODA) FROM THE INSHORE WATERS OF PORTO NOVO

Five species of *Acanthosquilla* have been reported to occur in the Indo-West Pacific Region (Manning[1]) of which *Acanthosquilla acanthocarpus* and *Acanthosquilla multifaciata* are known from the coastal waters of India (Kemp[2], Chhapgar and Sane[3] and Shanbhogue[4]) while Alikunhi[5,6,7], collected two larvae of *Acanthosquilla tigrina* from the plankton of the Madras coast and reared one of them in the laboratory to the adult stage.

The type specimen of *Acanthosquilla tigrina* (Nobili) lodged in Sarawak Museum (Kemp[2]), is a male measuring 45 mm and was found in Santubong, Borneo. Kemp[2] re-examined the specimen and supplemented Nobili's description with additional notes. Alikunhi[7] reared a 9·00 mm larva of *A. tigrina* in the laboratory. The larva lived for 81 days and underwent 8 moults before it metamorphosed into a young measuring 29·7 mm in length.

A male specimen of *Acanthosquilla tigrina* measuring 60 mm with right raptorial leg missing was collected on 4–8–1971 from the trawl catches of the inshore waters at Porto Novo (11° 29′ N–79° 48′ E). *Acanthosquilla tigrina* collected from Porto Novo waters agrees with the description by Kemp[2] especially in the presence of four pairs of fixed marginal teeth on the telson and with a posterior on the ventral margin of sixth abdominal somite but there are some differences. Firstly on the raptorial leg, the dactylus has 15 teeth (two terminal are partly damaged) but Kemp[2], mentions that in the type species the raptorial dactylus has 11 teeth and Alikunhi[7] found 13 teeth on the raptorial dactylus of the metamorphosed specimen. In *Acanthosquilla tigrina,* probably the number of teeth on the raptorial dactylus varies between 11 and 15 as seen in some other stomatopods, *Lysiosquilla maculata* and *Lysiosquilla spinosa* (Kemp[2]).

Secondly, on the telson, there are 15 denticles instead of 12 small denticles in between submedians as indicated by Kemp[2]. According to Alikunhi[7] there are 18 denticles in between submedians. The specimen is colourless in formalin. The differences noted above are minor variations from the previous descriptions of *A. tigrina* by Kemp[2] and Alikunhi[5,6,7].

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in Marine Biology, R. Natarajan.
Annamalai University,
Parangipettai 608 502, *September* 10, 1975.

---

1. Manning, R. B., *Proc. U.S. Nat. Mus.*, 1968, 124, No. 3641, 1.
2. Kemp, S., *Mem. Ind. Mus. Calcutta,* 1913, 4, 1.
3. Chhapgar, B. F. and Sane, S. R., *Jour. Biol. Sci. Bombay,* 1966, 9, 43.
4. Shanbhogue, S. L., *Bull. Centl. Mar. Fish. Res. Inst.*, 1969, 9, 35.
5. Alikunhi, K. H., *Curr. Sci.*, 1944, 13 (1), 18.
6. —, *Rec. Ind. Mus.*, Calcutta, 1952, 49 (3–4), 239.
7. —, *Proc. Symp. Crustacea,* Part II, 1967, p. 824.

FLUORESCENT ANTIBODY STAINING FOR DETECTION OF COCONUT ROOT WILT ANTIGEN

Fluorescent antibody (FA) technique serves as a useful tool for detecting *viral antigens* and their localization *in situ.* Coons *et al.*[1] developed this technique. Since then the technique has been widely employed for both plants and animals, infected with fungi, bacteria, viruses and mycoplasma[2–5]. Among plants considerable work has been done with atleast five viruses, namely, tobacco mosaic[6], wound tumor[7], southern bean mosaic[8], cauliflower mosaic[9] and narcissus yellow stripe[10].

The present report describes the use of FA technique in the detection of coconut root wilt antigen in the epidermal cells of tobacco. The coconut root wilt virus[11] was transmitted and maintained on *Nicotiana tabacum* cv. White Burley. The antigen was purified after Summanwar *et al.*[11]. The concentrated antigen was administered to rabbits weekly by four intravenous injections and two intramuscular injections (with Freund's adjuvant). The blood sample collected by bleeding the rabbits after 15 days following the last injection, was allowed to clot at room temperauture for four hours and stored overnight in a refrigerator for clot to shrink. The serum was clarified by low speed centrifugation at 4,000 rpm for 15 minutes. The antiserum reacted specifically with the coconut root wilt antigen and had a titre of 1: 1024.

Following Spendlove[12], the γ-globulins were precipitated with equal volume of 3·2·N ammonium sulphate. The precipitated globulins were recovered by centrifugation at 1,000 g for 30 minutes. The precipitate was dissolved in distilled water equal to the original volume of the serum and reprecipitated with ammonium sulphate as before. Residual sulphate was removed by overnight dialysis using distilled water. The pH of the globulin solution was raised to 9·0 using 0·5 M carbonate-bicarbonate buffer. 50 mg of fluorescein isothiocyanate (FITC) was added per gram protein in cold (4° C) and kept for 18 hours. The unconjugated dye was removed by overnight dialysis against buffer.

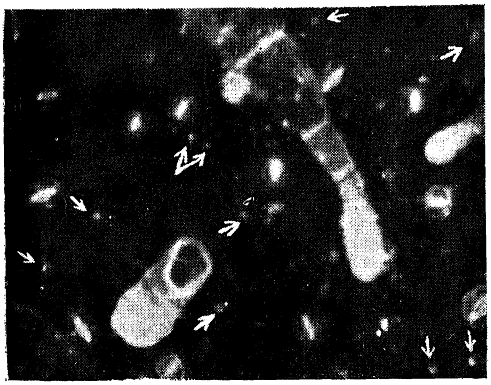

FIG 1. Diseased epidermal peel of *N. tabacum* cv. White Burley. Arrows indicate labelling of the coconut root wilt antigen with FITC.

Epidermal peels were taken from healthy and diseased tobacco leaves infiltrated in acetone. The peels were mounted on glass slides and allowed to almost dry. Labelled FITC was added to the glass slides and allowed to remain for 4-6 hours. The peels were then washed with buffered saline pH 7·5 (0·01 M PO_4 and 0·15 M NaCl) and mounted in the same buffer.

Observations were made using super-high-pressure mercury bulb HBO 200 with BG 3 and BG 12 light filters. Pictures were taken using suppression OG 1 filters. Healthy peels showed a uniformly dull orange fluorescence. On the other hand, the diseased peels showed an overall orange fluorescence, interspersed with deep apple green spots in the cytoplasm of the cells (Fig. 1, arrow marked—white spots). The deep apple green spots marked the labelling of the coconut root wilt antigen in the systemically infected tobacco epidermal cells.

The authors are thankful to Dr. S. P. Raychaudhuri, Head, Division of Mycology and Plant Pathology, Indian Agricultural Research Institute, New Delhi, for facilities and encouragement.

Division of Mycology and Plant Pathology, I.A.R.I., New Delhi 110012, August 25, 1975.

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1. Coons, A. H., Creech, H. J., Jones, R. N. and Berliner, E., *J. Immunol.*, 1942, **45** 159.
2. Kumar, D. and Patton, R. F., *Phytopathology*, 1964, **54**, 898.
3. Anger, J. G. and Shalla, T. A., *Ibid.*, 1975, **65**, 493.
4. Sinha, R. C., *Current Trends in Plant Pathology*, 1974, p. 99.
5. Kumar, D., Ghosh, S. K., Raychaudhuri, S. P., Nariani, T. K. and Varma, A., *Curr. Sci.*, 1975, **44**, 170.
6. Schramm, G. and Rottger, B., *Z. Naturforsch.* 1959, **14b**, 510.
7. Nagraj, A N., *Virology*, 1965, **25**, 133.
8. Worley, J. F. and Schneider, I R., *Phytopathology*, 1963, **53**, 1255.
9. Day, M. F. and Venablis, D G., *Australian J. Biol. Sci.*, 1961, **14**. 187.
10. Cremer, M. C. and Vander J. A. Velean, *Neth. J. Pl. Path.*, 1964, **72**, 105.
11. Summanwar, A. S., Raychaudhuri, S. P., Jagadish Chandra, K., Nam Prakash and Lal, S. B., *Curr. Sci.*, 1969, **38**, 208.
12. Spendlove, R. S., In *Methods in Virology*. (K. Maramorsch and H. Koprowski, Eds.) Vol. III, Academic Press, New York, 1967, p. 457.

ON THE USE OF MARINE CATFISH PITUITARY GLANDS FOR INDUCED SPAWNING OF SILVER CARP AND CATLA

THE Chinese silver carp, *Hypophthalmichthys molitrix* (Cuv. and Val.) and the Indian major carp, *Catla catla* (Ham.) were successfully induced spawned during 1962 and 1957 respectively using carp pituitary extract[1-4]. At present, the hypophysation technique is being used throughout India as a dependable method of pure carp seed production. An important problem in the intensive use of this technique, particularly along coastal areas, is the poor availability of carps for collection of pituitary glands. In order to overcome this limitation, studies on the use of marine catfish pituitary glands for induced spawning of cultivated carps have been conducted at this institution. During 1973, two species of Indian major carps, namely, *Labeo rohita* and *Cirrhinus mrigala* were induced spawned by injecting pituitary extract from two species of the marine catfish, *Tachysurus*[5]. In 1975, one more species of Indian major carp, *Catla catla,* and the Chinese silver carp, *Hypophthalmichthys molitrix,* were successfully induced to spawn by the same technique and these findings are reported in this communication.

The pituitary glands used for the experiments were collected from two species of marine catfish, *Tachysurus thalassinus* and *T. jella.* The glands were preserved in absolute alcohol and stored in a refrigerator. The donor catfishes were in the I or II stages of sexual maturity.

In all, four sets of silver carp and six sets of catla were treated with the marine catfish pituitary extract. All silver carp sets received the same dosage of pituitary extract of 40 mg/kg and 30 mg/kg body weight of fish for females and males respectively. Of these, two sets of silver carp responded and spawned. In the case of catla, three different dosages were tried. In two sets, the female fish received a total pituitary material of 40 mg/kg body weight and the male fish 30 mg/kg body weight. Both these sets spawned successfully. In three sets of catla, the female and male fish received a total pituitary dosage of 30 mg/kg body weight and 20 mg/kg body weight respectively. Of these three sets, two spawned successfully. One set of catla was injected at a low dose of 25 mg and 20 mg of pituitary gland per kg body weight of female and male fish respectively and this did not spawn.

The earlier observations on the successful breeding of rohu and mrigal[5] and the present similar findings on silver carp and catla confirm the utility of marine catfish pituitary glands for induced spawning of cultivated carps,

We are grateful to Prof. H. P. C. Shetty, Director of Instruction, College of Fisheries, for kindly going through the manuscript critically.

College of Fisheries, T. J. VARGHESE.
Univ. of Agricultural G. P. SATYANARAYANA RAO.
Sciences,
Mangalore 575 002, *August* 28, 1975.

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1. Alikunhi, K. H., *Proc. Indo-Pacific Fish. Coun. 10th Session,* 1963 *b,* p. 181.
2. —, Sukumaran, K. K. and Parameswaran, S., *Curr. Sci.,* 1963 *a,* **32,** 103.
3. Chaudhuri, H., *Ind. J. Fish.,* 1960, **7** (1), 20.
4. — and Alikunhi, K. H., *Curr. Sci.,* 1957, **26** (12), 381.
5. Varghese, T. J., Satyanarayana Rao, G. P., Devaraj, K. V. and Chandrasekhar, B., *Ibid.,* 1975, **44** (3), 75.

SOME ADDITIONS TO FUNGI OF INDIA

WE describe here three Fungi Imperfecti collected by us from forests surrounding Jabalpur (Central India). These are: *Trimmatostroma cordaicia* spec. nov. a new species and *Seimatosporium kriegerianum* (Bres). Morgan-Jones and Sutton and *Monodictys antiqua* (Corda) Hughes, two new fungus records for the country[1,3,4,8-10].

Trimmatostroma cordaicis spec. nov.

Colonies effuse, black, ellipsoid to irregular, large, easily detachable with the help of needle. Sporodochia pulvinate, sometimes, confluent, black. Mycelium immersed in the substratum, pale brown to dark brown, septate, extensively penetrating the host tissue. Stroma present, erumpent, brown, pseudoparenchymatous. Conidiophores semi-macronematous, meristematic, simple or sometimes loosely branched, short, straight or flexuous, closely packed together forming sporodochium, mid brown, smooth, 4–12 × 3–6 μ. Conidiogenous cells integrated, terminal roughly cylindrical, fragmenting. Conidia formed in irregular branches, basipetal chains, dry, at first simple later slightly verruculose, branched variously, lobed, clavate, irregularly subspherical, sometimes ellipsoid, dark brown to almost black, with 2–4 transverse septa and a few longitudinal and oblique septa, 8–20 × 8–15 μ (av. 16 × 8 μ).

Habitat : On dead wood, Adhartal, Jabalpur (M.P.), India, 20. VI. 1974, Leg. N. D. Sharma.

Type specimen has been deposited in herb. IMI, Kew, No. 183729, also in H.P.P., J.N.U., No. 17.

The genus *Trimmatostroma* have only two species *T. salicis* Corda and *T. betulinum* (Corda) Hughes[2]. The present species is close to *T. betulinum*, but differs from it in having slightly verruculose, dark brown to black conidia, having only few transverse and longitudinal septa. It is, therefore, accommodated under a new species.

The specific epithet derived from that of the eminent mycologist and founder of the genus *Trimmatostroma*, Professor A. C. J. Corda.

Trimmatostroma cordaicis spec. nov.

Colonia effusae, atrae, ellipsoidea vel irregulares, magnus. Sporodochia pulvinatus, nonnunquam confluentibus, atrae. Mycelium in substrata immersum, pallide brunneis vel atro-brunneis, septata. Stromata erumpentia, brunneis, pseudoparenchymatica. Conidiophora semi-macromemata, simplicia, coarctatus, pallide brunnea, levibus, 4–12 × 3–6 u. Cellae conidiogenae in conidiophoris incorporatae, terminales, plusminusve cylindrica, fragmentum. Conidia formis in rami irregulariter, basefuges catenata, aridus, ad primolevis, postea leviter verruculosa, ramosa varie, lobatus, clavata, irregulariter subsphaeroidea, nonnunquam ellipsoidea, atrobrunnea vel fusca, episporio crassiusculo, septis transversalibus 2–4 et septis longitudinalibus obliquisque paucis praedita, 8–20 × 8–15 μ (av. 16 × 8 μ).

Habitat : in lignum emortuis, Adhartal, Jabalpur (M.P.), India, 20, VI. 1974, Leg. N. D. Sharma.

Typus positus in Herb. I.M.I., Kew, No. 183729, also in H.P.P., J.N.O. sub numero 17.

Seimatosporium kriegerianum (Bres.) Morgan-Jones and Sutton, *Mycol. Pap.* 97, p. 37, 1964.

On leaves of *Callistemon lanceolatus*, Adhartal, Jabalpur (M.P.), India, August 1974, Leg. N. D. Sharma.

The specimen was referred to Dr. E. Punithalingam of the C.M.I., Kew, England, who remarked that : "This is an interesting collection which corresponds fairly closely with I.M.I. 163445 on *Callistemon palludosus* collected by Dr. Swart in Australia at present disposed in Herb I.M.I. as *Seimatosporium kriegerianum* (Bres.) Morgan-Jones and Sutton. This disposition should be regarded as tentative until Dr. Swart has completed his work on the group."

Sutton[6,7] confirming Shoemaker's[5] work in which the carlier name *Seimatosporium* Corda was taken up for those species previously referred to *Cryptostictis* Fuckel, transferred many species of latter to the former. The development of both the conidia and the conidiophores, the morphology of the conidia and their appendages, and the structure of the fructification conform quite well with the species of *Seimatosporium*. An unusual feature of the species is the development of the conidiophores. Apart from the normal annellophores described for several species of *Cryptostictis* (= *Seimatosporium*) *S. kriegerianum* shows a restricted type of sympodial development.

The specimen has been deposited in Herb. I.M.I., Kew, No. 189296 also in H.P.P., J.N.U., No. 18.

Monodictys antiqua (Corda) Hughes,
Canad. J. Botany, 36: 785, 1958.
= *Sporidesmium antiquum* Corda, Icon. Fung., 3: 4–1938 (PR).

On dead wood, Adhartal, Jabalpur (M.P.), India, Nov. 1973, Leg. N. D. Sharma.

On unpolished wood, Bohani, Narshingpur (M.P.), India, December 1973, Leg. N. D. Sharma.

Only three collections have been seen, but these were abundant and in excellent condition growing over rather dry wood. The present fungus closely resembles *M. antiqua* except for a few minor variations in the conidial measurements.

The specimens have been deposited in Herb. I.M.I., Kew, under Nos. 183730, 183733 and 183735 also in H.P.P., J.N.U., No. 19, 20 and 21.

The authors are grateful to Dr. G. P. Agarwal, Department of Post-Graduate Studies and Research in Botany, University of Jabalpur, Jabalpur, for helpful criticism and suggestions, and to Mr. A. Johnston, Director and Drs. M. B. Ellis and E. Punithalingam of the Commonwealth Mycological Institute, Kew, for help in the indentification of species.

Department of Mycology and Plant Pathology, J. N. Agricultural University, Jabalpur-4, M.P. — N. D. Sharma.
and
Department of Botany, N. E. S. Science College, Jabalpur, M.P., *August* 14, 1975. — S. R. Singh.

---

1. Butler, E. J. and Bisby, G. R., *The Fungi of India*, (Sci. Res. Monogr.), I.C.A.R., New Delhi, 1960, p. 552.
2. Ellis, M. B., *Dematiaceous Hyphomycetes*, Commonwealth Mycological Institute, Kew, Surrey, England, 1971, p. 608.
3. Mukerji, K. G. and Juneja, R. C., *Fungi of India* (1962–1972), Emkay Publ., Delhi, 1974, p. 223.
4. Rangaswami, G., Seshadri, V. S. and Lucy Channamma, K. A., *Fungi of South India*, Univ. Agric. Sci., Bangalore, 1970, p. 187.
5. Shoemaker, R. A., *Canad. J. Bot.*, 1964, **42**, 411.
6. Sutton, B. C., *Coelomycetes. II. Neobarclaya, Mycohypallage. Bleptosporium* and *Cryptostictis*, C.M.I., *Mycol. Pap.*, 1963, **88**, 50.
7. Sutton, B. C., *Coelomycetes. III. Annellolacinia* Gen. Nov., *Aristastoma, Phaeocytostroma, Seimatosporium* etc., C.M.I., *Mycol. Pap.*, 1964, **97**, 42.
8. Tandon, R. N. and Chandra, S., *Supplement to the List of Indian Fungi*, Univ. of Allahabad Studies, 1963, p. 246.
9. Tilak, S. T. and Rao, R., *Second Supplement to the Fungi of India*, Marathwada Univ., Aurangabad, 1970, p. 314.
10. Vasudeva, R. S., *Fungi of India, Supplement–I.* (Sci. Res. Monogr.), I.C.A.R., New Dehi, 1960, p. 206.

EFFECT OF ENVIRONMENTAL FLUCTUATIONS ON THE POLLEN STERILITY IN HOMO- AND HETEROZYGOTES OF RADISH (*RAPHANUS SATIVUS* L. VAR. *RADICOLA* PERS.)

IT has been proposed that the homozygous genotypes produced by inbreeding of an allogamous population differ from heterozygotes in response to environmental fluctuations during development.[1,2] Heterozygotes are, in general, developmentally more stable than homozygotes. However, there are few experimental data in support of this hypothesis. It is, therefore, desirable to know whether the same property of stability applies to pollen sterility. Hence a study has been made on the pollen sterility in three inbred lines of radish (*Raphanus sativus* L. Var. *radicola* Pers.) their F_1 hybrids and the original population in different period of the year.

Three highly homozygous inbred lines of radish, namely, LS–337/24 (I_{12}), LS–337/25 (I_{12}) and LS–43/51 (I_{16}), derived from the original population 'Saxa', and their F_1 hybrids have been used in the present investigation. These lines were selected from the genetic collection of Dr. S. I. Narbut of the Chair of Genetics and Plant breeding, Leningrad State University, Leningrad, and are cytogenetically well studied.[3–7] Mature flower buds from 5 plants in each form were fixed in a fresh mixture of 1 : 3 acetic acid and absolute alcohol three times during the flowering period, *i.e.*, in the beginning, in the middle and in the end of July in 1972. The temperature and the relative humidity prevailing on these dates were 32° C and 40%, 26° C and 60% and 21° C and 70% respectively. Same 5 plants were studied in all the three conditions of environments that prevailed on these dates. Anthers were stained and squashed in 1% acetocarmine. About 1000 pollen grains were examined in each plant. Percentage of abortive (sterile) pollen grains was calculated and the data were analyzed statistically by using both t and F tests.

Our results confirmed the previous results that inbred lines differ among themselves as well as from the original population in pollen sterility and that this difference, apparently, has a genotypic basis[4]. The present study shows that plants of the inbred lines, in contrast to the plants of the original population were more vulnerable to environmental fluctuations with regard to pollen sterility since the same plants were selected for study in all the three conditions of environment. Interestingly, the F_1 interlinear hybrids resembled the original population in this character. Besides, some hybrids, namely those obtained by crossing of LS–337/24 and LS–337/25, had practically negligible number of abortive pollen grains (less than 1%) and demonstrated heterosis. Other hybrids characteristically had low percentage of abortive pollen (Fig. 1).

From our results it follows that heterozygotes (population and F_1 interlinear hybrids) are genetically more balanced and possess better buffering properties than homozygotes (inbred lines). In other words, they display greater developmental homeostasis. Since the pollen abortiveness is known to depend largely on the behaviour of chromosomes during meiosis, an obvious conclusion would be that heterozygotes are more stable to environmental fluctuations in the chromosome behaviour as well than homozygotes. However, this needs experimental verification.

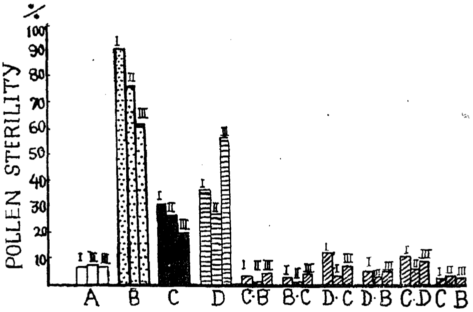

FIG. 1. Pollen sterility in the inbred lines of radish, their F_1 hybrids and the original population in different flowering periods of the year. A—Populalation; B—LS–337/25; C—LS–337/24; D—LS–43/51 and the reciprocal hybrids of these lines. I–32° C and 40% relative humidity; II–26° C temperature and 60% relative humidity and III–21° C temperature and 70% relative humidity.

The author expresses his sincere thanks to Prof. T. S. Fadeyeva and Dr. S. I. Narbut of the Chair of Genetics and Plant Breeding, Leningrad State University, Leningrad, for valuable suggestions and criticism.

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Patna University,
Patna 5 (Bihar), *August* 12, 1975.
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1. Lerner, I. M., *Genetic Homeostasis*, New York, 1954.
2. Mather, K., *Proc. Roy. Soc.*, *Ser. B*, 1955, **144**, 143.
3. Dayal, N., *Vestinik Leningr. Univ.*, *Ser. Biol.*, 1974, 3 (10), 142.
4. Dayal, N., *Ph.D. Thesis*. Leningrad State Univ., Leningrad, 1973.
5. Fadeyeva, T. S. and Dayal, N., *Genet. Issleyd.*, 1974, **5**, 103 (In Russian).
6. Narbut, S. I., *Genetika* (USSR), 1966, **5**, 89 (In Russian).
7. Narbut, S. I., *Vestnik Leningr. Univ.*, *Ser. Biol.*, 1967, **15** (3), 144 (In Russian).

DENDROPHTHOE FALCATA (LINN. F.) ETTING —A PARASITE ON THE LEAF OF *MANGIFERA INDICA* LINN.

Dendrophthoe falcata (Linn. f.) Etting belonging to the Loranthaceae is a partial stem parasite. It has been noted as a partial stem parasite on different forest host plants, like *Bombax ceiba* Linn.; *Grewia tilaefolia* Vahl; *Mangifera indica* Linn. etc. It has been noted here for the first time as a partial parasite on the leaf of *Mangifera indica* Linn. Here the parasite is attached to the midrib region. It produced flowers also (*see* photograph). The plant in comparison is delicate and is not gregareous as found on the stem. The parasite was noticed on a mango leaf in *Segvi* locality of Bulsar District. This adds a new data of its parasitism on leaf.

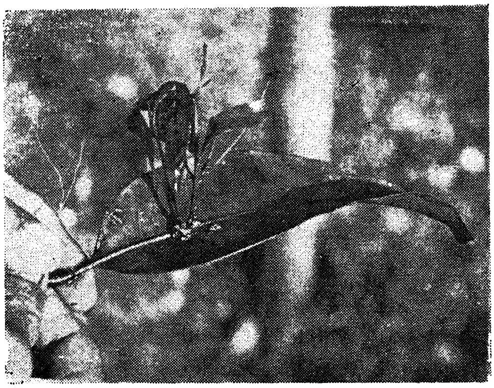

FIG. 1

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and
Department of Botany, Sardar Patel University, Vallabh Vidyanagar, Gujarat, *November* 21, 1975.

P. G. MORE.
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OCCURRENCE OF AN UNKNOWN WILD RACE OF *MANGIFERA* IN TRIPURA

IN view of the origin of the cultivated mango (*Mangifera indica* L.) somewhere in the North-Eastern India, in the Indo-Burma region[5], Tripura was thought to be one of the potential areas for collecting some useful germplasm for the mango breeding project at the IARI. The observations based on the survey of forest areas in May, 1975 are presented in this paper.

Habitat and Characteristic Features:

In the Southern parts of the Tripura State, about 120 kilometers from Agartala, a number of old mango seedlings were found growing wild on a hilly terrain on the bank of the river Gumti. The oldest of the seedlings appeared to be about a hundered year old and had an upright growth habit, rising to over 40 meters in hight. The trunk was almost straight and unbranched upto about 10 metres from the ground level. The trees were healthy and also free from mango 'malformation'—a serious disease threatening the mango industry in the North.

Leaves (Fig. 1): Glossy green; size very large (average lamina length 54·0 cm); shape elliptic-lanceolate; apex acute; margin slightly undulated; petiole very long (average length 11·0 cm), not thickened at the base; lateral nerves 31 pairs, prominent on both sides.

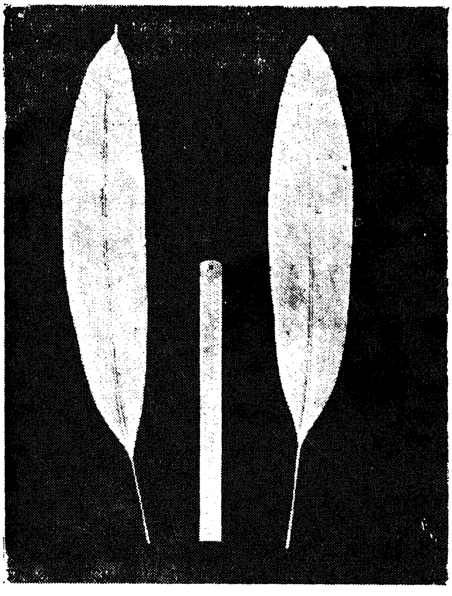

FIG. 1. Leaves of the unknown wild race of *Mangifera*.

Fruits (Fig. 2): Bearing profuse, in bunches of upto 50 fruits per panicle; size very small (average length 3·52 cm, breadth 2·52 cm and thickness 2·34 cm), average weight 12·5 gm; shape oblongish oval; shoulders unequal, ventral slightly more raised; back rounded; sinus absent; beak inconspicuous; apex rounded, skin light apricot-yellow, medium thick and smooth; glands indistinct and sparse; pulp lemon-yellow, very thin, soft, acidic and with scanty juice; stone ovalish-oblong (average length 3·40 cm, breadth 1·80 cm and thickness

1·00 cm), average weight 5·0 gm, veins thin and slightly sunken, fine fibres covering the stone.

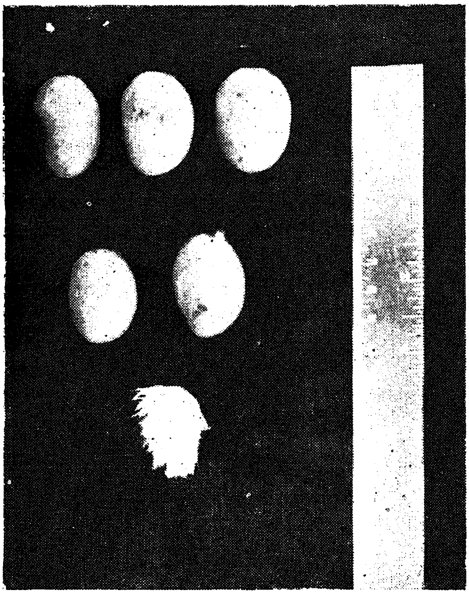

FIG. 2. A close-up of the fruits and stone of the type under description.

The description of this wild type does not appear to tally with any of the wild types of mango described in the literature[3,4]. The herbarium sheets of *Mangifera* species at the Indian Botanic Garden, Calcutta, also did not reveal a similar specimen.

It is reported that in the Section I of the genus *Mangifera* there are a number of species, other than *M. indica,* which have smaller fruits with very thin flesh[3]. However, excepting *M. sylvatica* Roxb. no other species of this section is found in the country and its fruits also differ with the present type, both in shape and size. Although some of the wild races of *M. indica* also bear small sized fruits, no detailed information is available regarding their fruit characters. The fruit size in the present case is nevertheless, much smaller than that of any of the 8 'Kalahandi' types of wild mango, descriptions of which are available in the literature[4]. The floral characteristics could not be studied, since the flowering was over. It would be of interest however, to compare its leaf characteristics with those of *Mangifera sylvatica* which has played an important role in the evolution of the cultivated mango.

According to Mukherjee[4], the leaves of the wild types of *M. indica* resemble with those of *M. sylvatica* which is found growing wild in the North-Eastern parts of the country. However, here again, there is no similarity, since the leaves of the latter (*cf.* Hooker's Flora of British India) are much smaller[2] (lamina 25–30 cm, petiole 5 cm or more) than the type described here (lamina 54 cm, petiole 11 cm). The number of lateral veins, considered to be characteristic of the species, is also much less (16–20 pairs) as compared to this wild type (31 pairs). Beside, the petiole is uniformly slender and not thickened at the base, as is the case with *M. sylvatica.*

The present observations suggest that the leaf characteristics, which are considered to be an important distinguishing feature (next only to the floral characters) for the different species in the genus *Mangifera*[1], show considerable difference from leaves of any of the species of *Mangifera* reported in the literature. Thus far, the largest leaf size (upto 36 cm) in *Mangifera* is reported in the species belonging to Section II of the genus[2]. None of the species of this section has so far been reported to occur in India. The available preliminary evidence indicates that this may, probably, be a new species or a species hybrid. However, further studies, especially of the floral characters, are required to arrive at a sound conclusion.

The case described here provides valuable material for the taxonomist for tracing the origin of the cultivated mango. The breeder may benefit if this wild type carries some useful genes like those for resistance to diseases and pests, self-fruit-fulness[6], and bearing in bunches.

The authors are grateful to Dr. R. N. Singh, Head of the Division of Horticulture and Fruit Technology, IARI, New Delhi and Mr. Mukul Sarkar, Director of Agriculture, Tripura, for providing the necessary facilities. Thanks are also due to Dr. R. B. Gosh, Systematic Botanist, Indian Botanic Garden, Calcutta.

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and
Directorate of Agriculture, Agartala (Tripura), — S. SEN CHOUDHURY.

August 22, 1975.

---

1. Mukherjee, S. K., *Bull. Bot. Soc. Bengal,* 1948, 2, 15.
2. —, *Lloydia,* 1949, 12, 73.
3. —, *Sci. and Cult.,* 1949, 15, 5.
4. —, *Ibid.,* 1950, 15, 469.
5. —, *Indian J. Hort.,* 1958, 15, 129.
6. Sharma, D. K. and Singh, R. N., *Hort Research,* 1970, 10, 108.

CLEFT GRAFTING IN *BURSERA PENICILLATA* (DC.) ENGL.

THE Linaloe tree of Mexico, *Bursera penicillata* (Moc. and Sesse ex DC.) Engl. (Syn. : *Bursera delpechiana* Poiss. ex Engl.) is a recent introduction to the economy of our country. Grafting experiments were started with a view to increasing the fruit yield of the species.

Bursera penicillata is dioecious and therefore it is obligatory that male and female trees should be in proximity to ensure pollination and fruit setting. R. W. Burton (1952) succeeded in budding female 'slips' to male trees. To graft a female bud on a male tree, is of little use for increasing the yield. On the contrary if male branches are grafted to female trees, the pollen produced will certainly facilitate easy pollination.

Therefore patch budding and cleft grafting of male scions were tried on a few female stocks at Coimbatore and Odanthurai. The patch budding gave poor results whereas cleft grafting was very successful.

Materials and Methods

Male branches of 2–3 cm. thickness were selected. Two slanting cuts were made on both sides. Female branches of the same thickness were selected and a cleft 3–5 cm. deep was cut through the centre on each branch. There was no pretreatment with hormones. The cut end of the male branch was inserted into the cleft and tied tightly with polythene ribbons and twine. The grafts established well within 3 to 4 weeks. The details of grafting carried out are as follows :

Place	Date of grafting	Number of clefts	Survival	Per cent of survival
Coimbatore	18–6–1973	5	5	100
Odanthurai, Coimbatore Central Forest Division	30–4–1974	10	6	60

The grafts established in 1973 at Coimbatore profusely flowered during this year and out of the 6 established grafts at Odanthurai 2 flowered.

The plantation at Odanthurai raised in 1967, has no male trees. Though the female trees, started flowering from the third year of planting there is no record of fruit set. There was very good fruit setting this year, in the female trees where the grafted male scion flowered and also in the adjoining female trees. This shows that pollination has taken place from the pollen of the male flowers produced by the grafted branch, thereby achieving the aim of this experiment.

Forest Research Centre, Coimbatore, Tamil Nadu, *August* 6, 1975.
A. M. MAHMOOD HUSAIN.
K. N. SUBRAMANIAN.
J. MADHAVAN NAYAR.

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1. Burton, R. W., *J. Bombay Nat. Hist. Soc.*, 1952, 152 (1), 116.

A NEW FRUIT ROT DISEASE OF *MALUS PUMILA*

DURING a survey of the local fruit market and store houses, a hitherto unrecorded, severe, soft rot disease of the apple (*Malus pumila*) fruit was observed. The disease manifested itself in the form of circular, light brown, water-soaked, soft areas on the skin of fruit which enlarged in diameter and depth as the disease advanced. Within a week, the whole fruit became soft and pulply (Fig. 1). The complete internal rotting was evident on cutting open the infected fruit.

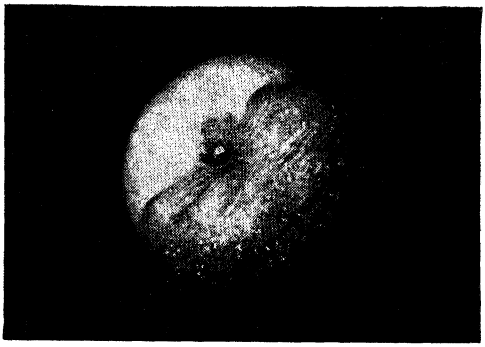

FIG. 1. Diseased apple fruit, infected with *Penicillium claviforme*, showing coremia on the surface.

The casual organism was isolated by routine pathological techniques and identified as *Penicillium claviforme* Bainier. The single spore cultures of the isolate were prepared. The pathogenicity of the fungus was established successfully by Koch's postulates. It was found that the pathogen invades the fruits through the injured surface. The fungus thrived best on PDA and host pulp agar.

The authors are grateful to the U.G.C., for providing financial assistance to one of us (T.S.T.).

Department of Botany,
University of Saugar,
Sagar 470 003 (M.P.),
January 2, 1976.
T. S. THIND.
S. B. SAKSENA.
S. C. AGRAWAL.

FIRST REPORT OF A TETRAPLOID SPECIES OF *CORCHORUS* FROM AFRICA

THE genus *Corchorus* (Tiliaceae) which includes two jute-fibre yielding species is spread throughout the tropics and subtropics. The total number of species is about 100[1] but the chromosome numbers of only 21 species are known[2–7]. The base number of all the species so far studied has been found to be $x = 7$. From the Indian subcontinent nine *Corchorus* species are reported of which the chromosome numbers of eight have been checked and all turned out to be diploids with $2n = 14$. Examination of five Australian *Corchorus* species (one also found in India, namely, *C. aestuans*) revealed the presence of a tetraploid species, *C. pascuorum*, the other species, namely, *sidoides*, *walcottii* and *elachocarpus*[7] being diploids. The chromosome picture of the *Corchorus* species of South America and the Carribbean islands is quite different. Of the five species checked for chromosome numbers, four, namely, *argutus*[5], *hirtus*[2], *orinocensis*[6] and *siliquosus*[2] were found to be tetraploids with $2n = 28$, the fifth *hirsutus*[7] being diploid. Chromosome numbers of ten African species (of which six are also found in India) were reported previously and all were diploids with $2n = 14$. This led us to believe that perhaps tetraploids are rare among African *Corchorus*. Recently however when we determined the chromosome number of *C. junodii* N. E. Brown, native to Mozambique, we found it to be a tetraploid with $2n = 28$. (Fig. 1).

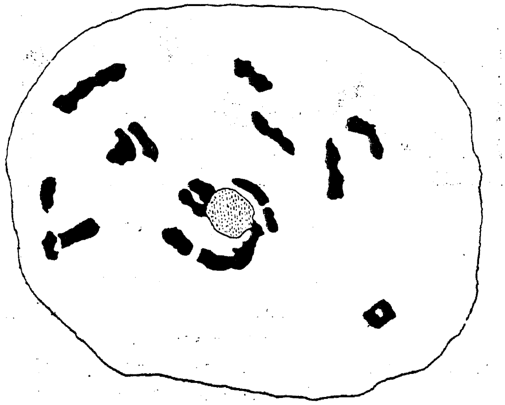

FIG. 1. Camera lucida drawing of a pollen-mother cell of *Corchorus junodii* at diakinesis showing 14 bivalents. [Note two bivalents attached to the nucleolus. × *c.* 600.

This is the first African tetraploid *Corchorus* species so far reported. It is a perennial plant like the two other tetraploid species of *Corchorus*, *siliquosus* and *pascuorum*. Meiosis in *C. junodii*, unlike that in *siliquosus* and *hirtus*, was slightly irregular with 40% pollen sterility. At metaphase I or diakinesis the 28 chromosomes were mostly present as 12 bivalents and one quadrivalent. From the present meiotic studies, it appears that it is a segmental allopolyploid. The voucher specimen is lodged in the herbarium of Botany Department, Dacca University, Bangladesh.

Thanks are due to Professor B. J. Harris of Ahmed Bellow University of Nigeria for arranging to obtain seeds of *C. junodii* from Mozambique.

Department of Botany, A. S. ISLAM.
University of Nairobi
and
Department of Botany, M. HAQUE.
University of Dacca, S. M. B. A. DEWAN.
Bangladesh, *September* 4, 1975.

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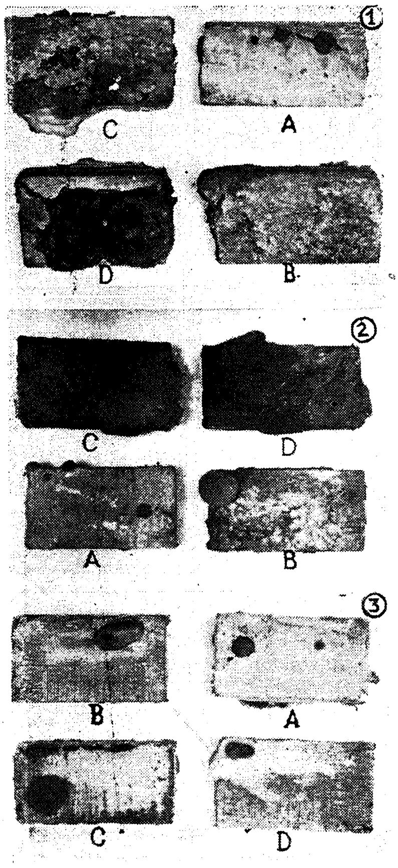
1. Willis, J. C., *A Dictionary of the Flowering Plants and Ferns*, Edited by Airy Shaw, H. K., Camb. Univ. Press, London, 1966, p. 282.
2. Islam, A. S. and Qaiyum, F., *Curr. Sci.*, 1961, **30**, 433.
3. Roy, K., *Curr. Sci.*, 1962, **31**, 205.
4. Datta, R. M., Panda, B. S., Roy, K., Bose, M. M. and De, T. K., *Bot. Magazine*, 1966, **79**, 939.
5. Love, A., IOBP chromosome number reports up to 1974 in *Taxon*, 1957–1974.
6. Cristobal, C. L., *Kurtziana*, 1967, **4**, 142.
7. Determined in the principal author's laboratory.

INDUCTION OF BASIDIOCARP FORMATION OF THREE STRAINS OF *FOMES DURISSIMUS* LLOYD IN CULTURE

INDUCTION of basidiocarp formation of fungi in culture still remains a great problem as they vary widely in their responses towards different natural conditions. No single and standard method has yet been devised that may be applicable in all cases. The literature in this field of research is, however, comparatively meagre[1–8]. The three strains of *Fomes durissimus* growing extensively on trees of great economic value, *Swietenia mahogani*, *Casuarina equisetifolia* and *Mimusops elengi* cause considerable amount of decay annually. No work has been done so far on the production of fruit bodies of *F. durissimus* in culture. The present experiment is an attempt to induce fructifications of three strains of *F. durissimus* Lloyd in culture.

At first, attempts were made with different nutrient agar media in Petridishes which were subjected to various conditions of moisture, light and temperature in the laboratory. But none of the strains of the test fungus produced typical fruit bodies in any of the media in spite of sufficient vegetative growth in all the cases and the formations of "anlagen" only in malt-agar medium. In about 60 days, several mycelial knots (anlagen) were found to develop sparcely near the lid of the Petridish. At this stage, the lid was carefully removed and the cultures kept inside a bell-jar under high humidity. Some of these knots gradually increased in size and after four months, looked like small knobs with deep brown colour. An experiment was then started to induce the three aforesaid strains of the test-fungus to fructify on wood blocks of their respective hosts. Sapwood blocks ($2'' \times 1'' \times \frac{1}{3}''$) were sterilized, aseptically introduced into Kolle flasks, four in each over a 30 days old culture of the respective strains of the test-fungus on 2% malt-agar slants. The wood-blocks were vigorously attacked on all sides by luxuriant growth of the mycelium of respective strains of *F. durissimus* within one month; after two months they were completely covered by thick, tough, brown coloured mat with a few mycelial 'knots' at places, on the surface of the wood blocks. Some of these 'knots' slowly increased in size and after four months looked like small 'knobs', more or less rounded in shape and dull yellow in colour. The flasks were then kept under diffused light and sterilized water was sprayed over the blocks at intervals. In about eight months the fruit bodies became hard, woody and dark brown in colour (Figs. 1–3). Sections through the hymenial surface revealed the presence of regular pore tubes with well developed basidia and basidiospores and their measurements agreed closely with those of the fruit-bodies occurring in nature. From the foregoing, it is clear that for the induction of typical fruit bodies of the three strains of *F. durissimus* the following conditions are essential: (i) sufficient supply of nutrients; (ii) moist atmosphere; (iii) favourable temperature and light. This is in conformity with the observations made earlier by Banerjee and Sinha[6].



FIGS. 1–3. Fig. 1. Fructifications of the strain of *Fomes durissimus* isolated from *Swietenia mahogani* at different stages of development in wood-block-cultures. Fig 2. Fructifications of the strain of *F. durissimus* isolated from *Casuarina equisetifolia* at different stages of development in wood-block-cultures. Fig. 3. Fructifications of the strain of *F. durissimus* isolated from *Mimusops elengi* at different stages of development in wood-block-cultures.

Department of Botany, S. SANTRA.
University of Burdwan, B. NANDI.
Burdwan, West Bengal,
October 30, 1975.

1. Long, W. H. and Harsch, R. M., *J. Agric. Res.*, 1918, **12**, 33.
2. Bose, S. R., *Bot. Gaz.*, 1929, **87**, 665.
3. Venkatarayan, S. V., *Phytopath.*, 1936, **26**, 153.
4. Badcock, E. C., *Trans. Brit. Mycol. Soc.*, 1941, **25**, 200.
5. —, *Ibid.*, 1943, **26**, 127.
6. Banerjee, S. N. and Sinha, A. K., *Sci. and Cult.*, 1955, **20**, 612.
7. — and Sarkar, A., *Indian J. Mycol. Res.*, **2**, 80.
8. — and Nandi, B., *Ibid.*, **7**, 39.

A REPORT OF LEAF SPOT DISEASES ON SOME VEGETABLE, FODDER AND ORNAMENTAL PLANTS

A NUMBER of leaf spot infections were observed on vegetable, fodder and ornamental plants near Guntur, (India), during the rainy season and winter of 1974–75. Fruit rot of bhendi and leaf spots on *Amarantus viridis* L., *Ipomea sepiaria* Koen, *Nyctanthes arbortrists* L., *Chrysanthemum cinerariafolium* Vis, which are new records for India, are reported here.

The following procedure was adopted for all the diseases. The pathogenic fungus was isolated on Czapek–Dox agar medium from several infected regions. The pathogenecity of the fungus isolated from diseased areas was proved by spraying the healthy plants with spore suspension, prepared in sterillized water from one week old culture and covering the plants with moist polyethylene bags for three days at room temperature (27–31° C). Reisolations yielded the original fungus in all the tests. The cultures were sent to CMI, England, for confirmation of the identity of the pathogens. The details of the disease symptoms and the characteristics of the pathogens concerned are given in Table I. The fruit rot of bhendi and the leaf spot

TABLE I

Disease symptoms, cultural characters of the pathogens and their identity

Host	Disease	Symptoms	Cultural characters	Identity of the pathogen
Abelmoschus esculentus (L.) Moench (Bhendi)	Fruit rot	Blackening of tips of developing fruits resulting in progressive drying from tips to base; small black necrotic spots are present on the fruits	Mycelium green to bluish green, cottony, conidia in chains of 5–8, measure 38 μ–70 μ × 8 μ–14 μ.	*Alternaria* state of *Pleospora infectoria* Fuckel. (IMI 194871).
Amarantus viridis L.	Leaf spot	Small, brown to black necrotic lesions (3–4 mm)	Mycelium black, conidiophores neither septate, nor branched, conidia single celled, cylindrical with few vacuoles	*Clletotrichum* sp. Atypical isolate belonging to *C. dematium* (Pers. ex. Fr). Grove group (IMI 194869).
Ipomea sepiaria Koen	Leaf spot	Brown, circular necrotic lesions with chlorotic halo (0·5–1·0 cm). Necrotic zone showed typical concentric rings	Mycelium green to bluish green, cottony, conidia in chains of 6–8, measure 35 μ–70 μ × 8 μ– 14 μ.	*Alternaria* state of *Pleospora infectoria* Fuckel. (IMI 194873).
Nyctanthes arbor-tristis L. (Har Singar).	Leaf spot	Large, oblong, brown, necrotic lesions with yellow borders (1–2 cm × 0·5–0·8 cm) both along the margin and on the lamina, necrotic lesions gradually increased in size covering entire lamina	Mycelium white turning to ash grey, condiophores hyalline bearing conidia apically, conidia unicellular, hyaline, cylindrical with few vacuoles, measure 9 μ–15 μ × 3 μ–4 μ, setose processes absent	Conidial stage of *Glomerella cingulata* (Stonem) Spauld and Schrenk (IMI 194870).
Chrysanthemum cinerariafolium Vis.	Leaf spot	Large, dark brown, necrotic lesions mostly along the margins. The disease spreads from basal to upper leaves	A pycnidial fungus, mycelium brown, pycnidia numerous, immersed, 15 μ–25 μ, pycnidiospores hyaline, oval, single celled, measure 4–5 μ × 2–3 μ	*Phoma herbarum* Westend (IMI 194868). Since the pathogen is a soil fungus it is presumed that spores from soil may be splashed by rain to the basal leaves causing leaf spots, from which the disease spreads to upper leaves.

of Chrysanthemum had developed extensively and appear to be of economic importance to the cultivation of the respective hosts.

Grateful thanks are due to Dr. A. Johnston, Director, CMI, England, for forwarding the reports on cultures sent to CMI, for identification. We are thankful to Sri B. V. Subbiah, JRF, Department of Botany, for his help in this work. One of us (CSKV) is thankful to CSIR, New Delhi, for the award of Junior Research Fellowship.

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September 17, 1975.

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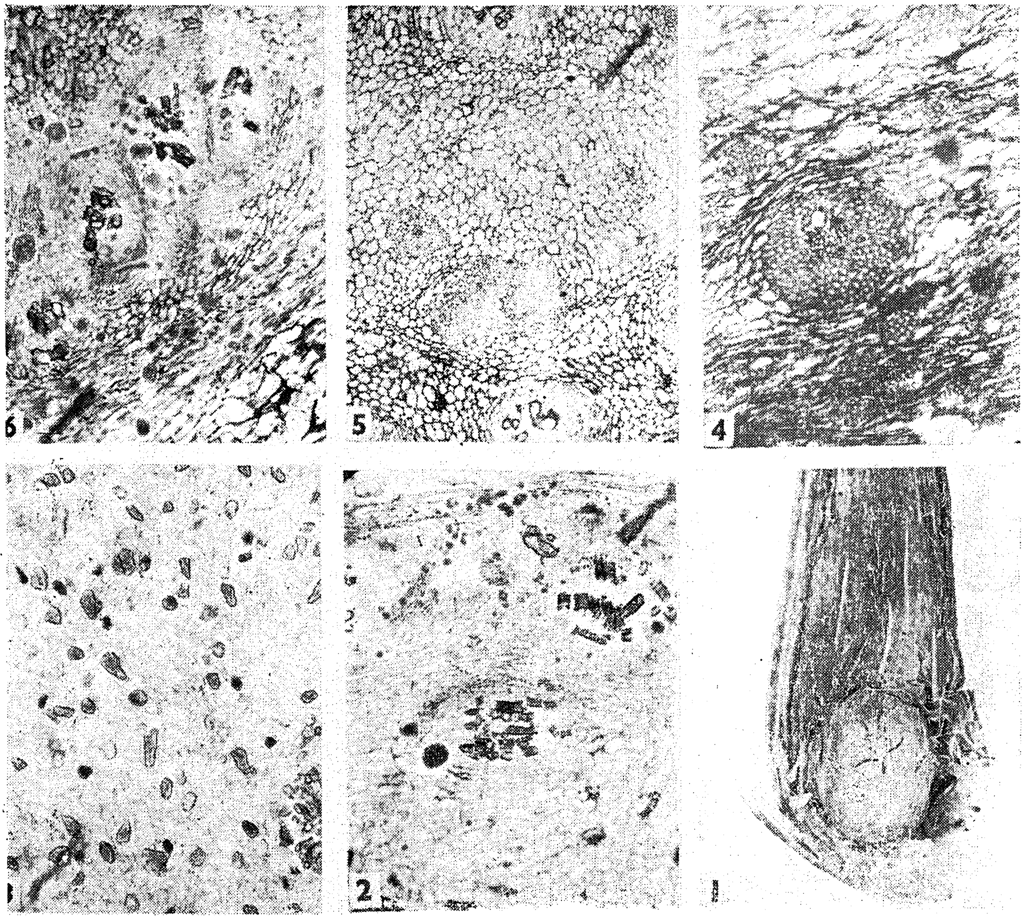
A. S. RAO.

ANATOMICAL STUDIES ON THE BULBILS OF COCONUT

BULBILS of different origin have been reported earlier in coconut[1-4]. However, the anatomy has not been investigated so far. The abnormal tissue described below initiated the present study.

A thick fleshy globular tissue (diameter = 3·8 cm) was observed within one of the leaf bases of a palm (Fig. 1), near the Calicut University campus. The material was preserved in FAA and the tissue showed cellular structure with many vascular bundles oriented irregularly and the parenchymatous cells were isodiametrical with the peripheral layers compressed (Fig. 2). The neighbouring leaf base fibres and tissues around this structure adhered to its surface. Internal tissue showed the presence of tanniferous cells distributed all over and the globular structure was devoid of fibrous tissue unlike the normal leaf base (Fig. 3). In the normal tissue of the leaf base highly thickened sclerenchymatous patches were profusely distributed (Fig. 4). Regions of actively dividing parenchymatous tissues were also seen scattered within the tissue and some of them contained differentiated vascular elements (Fig. 5). This indicated differentiation within the tissue. Further, the surface of the abnormal tissue was not smooth, instead, the sections showed undulated periphery, as represented by rudimentary patches of cells subtended with a few closely packed vascular bundles representing axillary structures (Fig. 6).



Figs. 1–5. Fig. 1. L.s. of leaf base showing the abnormal tissue. Fig. 2. Cellular structure and distribution of vascular bundles in the abnormal tissue. Fig. 3. Internally distributed tanniferous cells in the tissue. Fig. 4. Profusely distributed sclerenchymatous patches (fibres) in the normal tissue in leaf base. Fig. 5. Actively dividing and differentiating meristematic tissues in the abnormal structure and Fig. 6. Peripheral tissue subtended with closely placed vascular traces of axillary structures.

It may be surmised that this structure might probably be an axillary shoot/inflorescence which might have got included within the basal tissues of the leaf base during ontogenesis, as a result of certain dishormony in growth, the etiology of which remains unknown. However, such abnormal situations are extremely rare and might be teratological.

Department of Postgraduate Studies and Research in Botany,
University of Calicut,
Kerala, *September* 29, 1975.

C. Mohandas.
P. T. Annie.
K. Pavithran.

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1. Davis, T. A., *J. Bombay Nat. Hist. Soc.*, 1948 *a*, 47, 527.
2. —, *Bull. Indian Centr. Coconut. Cttee.*, 1954, 7, 202.
3. —, *Oleagineux*, 1967, 22, 19.
4. Furtado, C. X. and Barreto, P. C., *Poona Agric. Coll. Mag.*, 1920, 12, 81.

SHORT SCIENTIFIC NOTES

Pre-historic Fauna of Kalibangan, Rajasthan, India

While studying the collection of animal remains of Kalibangan, Ganganagar District, Rajasthan excavated by the Archaeological Survey of India during 1962–65, a few interesting skeletal fragments came to our notice. The age of these fragments is about 3500 to 4000 years. Based on 1500 fragments of different bones, we have identified 18 species. Since a detailed report on the collection will take more time a preliminary account is given in this note. The total numbers of fragments of different bones of the species identified are given in the parenthesis.

TABLE I

Phylum	Class	Order	Family	Species
Chordata	Mammalia	Carnivora	Canidae	*Canis familiaris* Linn. (8) *Canis lupus* Linn. (8) *Felis chaus Goldust* (38)
		Proboscidea	Elephantidae	*Elephas maximus* Linn. (1)
		Perissodactyla	Rhinoceretidae Equidae	*Rhinoceros unicornis* Linn. (6) *Equus hemionus* Pallas (1)
		Artiodactyla	Svidae Camellidae Cervidae	*Sus scrofa cristatus* Wagner (68) *Camleus* sp. (8) *Axis axis* Erxl. (1) *Cervus unicolor* Kerr (1) *Cervus duvauceli* Cuv (16)
			Bovidae	*Bos indicus* Linn. (83) *Bubalus bubalis* Linn. (149) *Capra hircus aegagrus* Erxl. (45) *Ovis orientalis vignei* Blyth (94)
	Aves	Columbiformis Galliformis	Columbidae Phasianidae	*Columba livia* Gmelin (2) *Gallus sonneratti* Term. (1)
	Reptilia	Chelonia	Testudinidae	*Batagura baska* Gray (2) *Kachuga tectum* Gray (2) *Chitra indica* Gray (18) *Lissemys punctata* (Bonnaterre) (1)

It may be stated that Banerjee and Chakraborty (1973) have already reported one horned Rhinoceros from this collection. Nath (1969), in connection with the study of the role of animal remains in the early prehistoric cultures of India, casually mentioned about some of the common names of domestic and wild animals. The occurrence of the species mentioned by us in Rajasthan and in Harappa (Prasad, B., 1936) and Mohenjodaro (Sewell, R. B. S. and Guha, B. S., 1931) prove their extensive distribution in North Western India in the early days. This confirms the geological evidence that this arid zone area is of recent origin.

Thanks are due to Dr. S. Khera, and Dr. K. K. Tiwari, Deputy Directors of our Survey, for the facilities provided.

Zoological Survey of India, Calcutta 700 016, *August* 28, 1975. S. BANERJEE. R. N. MUKHERJEE.

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1. Banerjee, S. and Chakravorty, S., *Sci. and Cult.*, Calcutta, 1973, 39, 430.
2. Nath, B., *Indian Mus. Bull.*, Calcutta, 1969, 4, 107.
3. Ellerman, J. R., Morrison and Scott, T. C. S., *Cheklist of Palaearctic and India Mammals*, 1758 to 1946, 1951. p. 1. British Museum (Natural History) Chromwell Road, S. W. 7, London.

Freshwater Cladocera (Arthropoda: Crustacea) from Northeast India

Very little is known about cladoceran fauna of Northeast India. Cladocera from this area during the past year, were collected from freshwater bodies of some areas of Meghalaya and Manipur. These collections reveal 17 species of which 14 are new records from the region and 3 are new to India. A list of the species along with collection localities for each is given below, new records from India being marked by (*). The seventeen species recorded below belong to five families and thirteen genera.

Sl. No.	Cladocera : Species	Collection locality
	Family: SIDIDAE	
1.	*Diaphanosoma sarsi* Richard	.. Ward lake, Shillong (Meghalaya).
	Family: DAPHNIDAE	
2.	**Simocephalus exspinosus* (Koch)	.. Ward lake, Shillong (Meghalaya).
3.	*Simocephalus elizabethae* (King)	.. Ward lake, Shillong (Meghalaya), Ninthoukhong (Manipur).
4.	*Scapholeberis kingi* Sars	.. Ward lake, Fruit Garden, Shillong (Meghalaya).
5.	*Ceriodaphnia rigaudi* Richard	.. Ward lake, Shillong (Meghalaya).
6.	*Moina brachiata* (Jurine)	.. Ward lake, Fruit Garden, Shillong (Meghalaya).
	Family: BOSMINIDAE	
7.	*Bosmina longirostris* (O.F. Muller)	.. Fruit Garden, Shillong (Meghalaya).
	Family: MACROTHRICIDAE	
8.	*Streblocerus serricaudatus* (Fischer)	.. Kangvai (Manipur).
9.	*Macrothrix spinosa* King	.. Sendra (Manipur)
10.	*Ilyocryptus halyi* Brady	.. Golflinks, Shillong (Meghalaya).
	Family: CHYDORIDAE Sub-family: CHYDORINAE	
11.	**Camptocercus rectirostris* Schødler	.. Lady Hydari Park, Shillong (Meghalaya).
12.	**Acroperus harpae* Baird	.. Ward lake, Lady Hydari Park, Shillong (Meghalaya).
13.	*Alona affinis* (Leyaig)	.. Fruit Garden, Mawlai, Shillong (Meghalaya).
14.	*Alona rectangula* Sars	.. Kangvai (Manipur).
15.	*Chydorus globosus* Baird	.. Ward lake, Shillong (Meghalaya).
16.	*Chydorus sphaericus* (O.F. Muller)	.. Ward lake, Shillong (Meghalaya).
17.	*Chydorus denticulatus* Henry	.. Lady Hydari Park, Shillong (Meghalaya).

The author is thankful to the Director, Zoological Survey of India, Calcutta, for providing the facilities to work out these collections and to Dr. A. K. Ghosh, Officer-in-Charge, Eastern Regional Station, Zoological Survey of India, Shillong, for his valuable suggestions and guidance.

Eastern Regional Station, S. G. Patil.
Zoological Survey of India,
Shillong 793 003, *November* 20, 1975.

A New Leaf Spot Disease of *Plumeria acutifolia* Caused by *Alternaria alternata*

A severe leaf spot disease of *Plumeria acutifolia* a popular ornamental plant, was observed during the rainy season of 1975 around Guntur, Andhra Pradesh. The disease manifests itself in the form of brown necrotic spots measuring 1–4 mm. The spots appear all over the leaf surface. The young as well as mature leaves were seen to be equally attacked.

Pure culture of the fungus was obtained on PDA medium. The pathogenicity was tested on detached leaves. The typical symptoms developed after 5–6 days. The hypae are septate and branched, with smooth walled conidia occurring in chains, 2–5 transverse and 1–2 longitudinal septa are present. The conidia measures 24–34 $\mu \times$ 8–16 μ. The causal organism was identified and confirmed as *Alternaria alternata* (Fr) Keissler (IMI 197735).

Grateful thanks are due to Dr. Ellis, CMI, England, for the identification of the fungus.

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December 15, 1975.

Role of Tulsi Plant (*Ocimum sanctum*) in Control of Mango Fruitfly, *Dacus correctus* Bezzi (Tephritidae: Diptera)

Occurrence of fruitfly, *Dacus correctus* Bezzi on Mango and chiku in South Gujarat has been reported by Shah and Vora (1974). In working out the extent and the time of incidence as well as the damage to Mango and chiku crop, it was found for the first time that tulsi plant, *Ocimum sanctum* worked as an attractant for the fruitfly in the area. Observations on the population counts revealed that all the fruitflies attracted to tulsi plants were males only. On chemical analysis it was found that the leaves contained 20·4% methyl eugenol in its total essential oil content, and thus, played a role in attracting male fruitflies as sex attractant. Preliminary biological experiments confirmed that methyl eugenol would act as a bait material (Sex attractant) for the male flies. This behaviour is taken advantage of in the studies of population density of fruitflies in the garden.

Gujarat Agricultural University, A. H. SHAH.
N. M. College of Agriculture, R. C. PATEL.
Navsari Campus, *December* 11, 1975.

---

1. Shah, A. H. and Vora, J. V., 1974 "Occurrence of *Dacus correctus* Bezzi (Tephritidae : Diptera) on Mango and chiku in South Gujarat," *Ind. J. Ent.*, 1974, 36 (1),

Polygonum dumetorum **L. A New Host for** ***Puccinia polygoni-ambibii*** **Pers.**

Polygonum dumetorum L. a wild growing plant in Kashmir between 6,000–9,000 ft was observed severely attacked by *Puccinia polygoni-ambibii* Pers. collected during August–September 1975.

The disease manifests itself in the form of irregular brown patches measuring 2–5 mm across, found only on dorsal side of the leaf. Average patches of the disease per leaf was found between 10–15, only mature leaves were seem to be attacked.

The fungus is identified as *Puccinia polygoni-ambibii* Pers. (IMI 197021). The present fungus has not been recorded on the above host and hence constitutes a new host record.

The author wishes his sincere thanks to Dr. M. B. Ellis and Dr. Mordue, CMI Ferry Lane, Kew, Surrey, England, for identification of fungus and to Dr. G. N. Javeid, Reader, Department of Botany, for providing the facilities.

Department of Botany, A. H. MUNSHI.
University of Kashmir,
Srinagar 6, *November* 14, 1975.

Alkaloid Distribution in the Fruit of *Solanum khasianum*

The variability in the steroid content in *Solanum khasianum* has been attributed to various factors. Saini *et al.*[1] and Chandra *et al.*[2] found maximum amount of steroid at a particular stage of fruit harvest. Khanna and Murty[3], however, reported that the plant maturity and the stage of fruit harvest at a particular period had an influence on the solasodine build up. Tht present note deals with the study of correlation of some characters with the steriod content.

Seeds of *S. khasianum* obtained from various sources were sown during October–November, 1967 and the seedlings transplanted in the field during January–February, 1968, at a distance of 90 cm. in rows 90 cm. apart. A number of plants were chosen at random and the data were collected on fruit size, number of seed per fruit and solasodine per cent. Solasodine was determined as described earlier[3], from fruits at uniform stage of development. The correlation coefficient between solasodine content and other variables has been calculated and presented in Table I.

TABLE I

Correlation coefficient between solasodine content and other characters in Solanum khasianum

Characters	Correlation coefficient
1. Fruit size and seed number	+0·90
2. Fruit size and alkaloid	+0·21
3. Weight of the fruit and akaloid	+0·12
4. Volume of the fruit and alkaloid	+0·13
5. Relative density of fruit and alkaloid	−0·15

It was pointed out[4], that *g*-alkaloid is localised to the gelatinous layer covering seed surface. In the present experiment the seed number is correlated with fruit size, but no correlation was observed between the *g*-alkaloid content and fruit size.

Janaki Ammal and Bhat[5] also observed increased *g*-alkaloid content in induced polyploids but with a reduced seed number. Thus it appears that the mucillage secreting cells responsible, for the *g*-alkaloid are not only confined to the seed coat but also to other parts of the fruit.

National Botanic Gardens, ANDRA S. MURTY.
Lucknow, *October* 27, 1975.

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1. Saini, A. D., Mukherjee, M. and Biswas, R. C., *Ind. J. Plant Physiol.*, 1965, **8**, 103.
2. Chandra, V., Basant Singh, Singh, A. and Kapoor, L. D., *Indian Forester*, 1970, **96**, 352.
3. Khanna, K. R. and Murty, A. S., *Planta Medica*, 1972, 21, 182.
4. Saini, A. D., *Curr. Sci.*, 1966, 35, 600.
5. Janaki Ammal, E. K. and Bharati Bhatt, *Curr. Sci.*, 1971, **40** (3), 70.

REVIEWS AND NOTICES OF BOOKS

An Introduction to Tensor Analysis. By John R. Tyldesley. (Longman Group, London), 1975, Pp. xii + 114. Price £ 3.50.

The author's stated purpose is to introduce tensors through examples in physical sciences, but the book could as well have been titled as 'tensor analysis theory and applications to mechanics of continua'. The first and fifth chapters treat tensors in cartesian and generalised coordinates respectively. The second, third and fourth chapters can be read as applications of tensors to continuum mechanics and to a few topics in field theory. Incidentally it may be mentioned that there are several books, both elementary and at an advanced level, precisely addressed to these topics.

The treatment of employing tensors to read an equation or write it in concise form is unnecessarily detailed, but detailed treatment where necessary is lacking. For example, while treating tensor calculus there are no examples on partial differentiation; integration is explained by one example without specifying that it is a line integral—more examples on surface and volume integrals would have been in order. The vector notations like div, grad, curl have not been extended to higher order tensors. In summary the book is not well organized and lacks adequate treatment of important concepts which should be conveyed to an applied scientist or an engineer in a simple manner.

C. Uberoi.

Introductory Quantum Chemistry. By A. K. Chandra. (Tata McGraw-Hill Publishing Co. Ltd., New Delhi), 1974. Pp. viii + 227. Price Rs. 28·50.

It is much easier to write a book on Quantum Mechanics than one on Quantum Chemistry. Quantum Chemistry is a very vast subject, and proper justices cannot be done in one single book. Most of the books therefore follow a pattern which heavily shows authors' bias. The present book is no exception. I would have expected some discussion at least of electrical and magnetic properties of molecules and intermolecular forces. Any phenomenon which cannot be explained by classical physics should come under Quantum Chemistry.

It must be stated at the very outset that up-till now the best book on Quantum Chemistry is the book by Eyring, Walter and Kimble. But that book is rather heavy going, for chemistry students of Indian Universities. Chandra has tried to follow the Mathematical rigour of Eyring's book upto a certain stage. In so doing, however, he has omitted certain points which are rather unfortunate. He has not given discussion on symmetry operators and their commutation relation with Hamiltonian. This has necessitated a crude algebraical treatment of Huckel molecular Orbital. In addition, he hts used these operators in explaining Woodward-Hoffman rule, which will carry no sense to the students. The author has introduced the terms selection rule, long before he has treated transition probability. This is neither permissible nor desirable.

One exceptional feature of this book is a large number of worked out problems. The problems are very well selected and if the students are made to work them out, they will get a good grounding of the subject.

On the whole the author has tried to give a modern outlook to the subject and I am sure the students in Indian Universities will benefit by reading this book, if helped and supplemented by the teachers.

S. Basu.

Man-made Lakes and Human Health. Edited by N. F. Stanley and M. P. Alpers. (Academic Press, London, New York), 1974. Pp. xvi + 495. Price £ 12.40.

This is a very timely publication, to focus the attention of the Engineers and the Governments engaged in the construction of big dams, on the 'Health hazards of Man-made lakes' and to emphasize the vital importance of conducting ecological studies with special reference to the Health problems of the geographical area concerned, much prior to the building of the dam itself.

Man-made lakes are all examples of human interference with eco-systems that have evolved slowly but are now subject to profound disturbance. Though, the increased agriculture and power potential created by dams should lead to increased health and well being, in the human populations living around them, millions of people are suffering today from infectious diseases associated with the development of Man-made lakes

Many of these diseases have not yet been controlled particularly schistosomiasis, malaria and arbovirus infections.

The justification for building the dam in the first place is based on economic grounds and human health is frequently forgotten. Failure to attend to the health aspects of any large Man-made lake may lead to economic disaster. For this to be arvested, health planning must be built into the project from its inception, long before anything is build and continued long after the lake has filled, until the health provisions are fully incorporated into the public health structure of the country concerned.

This volume vividly portrays, with an in-depth study, the health problems posed by the construction of nine lakes in different regions of the world. The specific geographical areas exemplified are: The Kariba dam over the river Zambesi in Central Africa, the Aswan High dam and the resulting Lake Nasser in Egypt, the Ord river dam of tropical Australia, Ubolratna dam complex of Thailand, reservoirs of the Great Central California basin, the Brokopondo research project in Netherland, the Kisumu project in Kenya, the Kainji dam in Nigeria and Volta lake of Ghana. Besides the engineering problems, the ecological disturbances caused, the health hazards encountered during and after the construction of the dams, the lessons to be learnt from the lack of perspective planning and tragic events that occurred in the resettlement of the population in some projects have been brought out very graphically.

The health hazards associated with the specific projects dealt with in detail include schistosominiasis, malaria, arbovirus infections, trypanosomiasis, filariasis, leptospirosis, leishmaniasis, ankylostomiasis plus malnutrition and other facets of parasiticism that determine the destiny of man particularly in the developing countries of the world.

Evidence presented in the volume indicate that dams and Man-made lakes may descrease as well as increase human health and prosperity; planning should be multidimensional from the beginning and be based, where possible, on specific local data; ecological changes following the construction of the dam should be monitered; engineers, economists and scientists should co-ordinate their activities at all stages and human health must be given explicit consideration at all stages of the planning, construction and use of Man-made lakes.

With its wealth of technical details and in-depth studies, this volume will prove extremely useful as a reference work for engineers, public health personnel, agriculturalists, administrators and students of the biomedical sciences and provides sufficient accurate data to enable governments, public health authorities and the scientific community to make intelligent decisions about Man-made lakes.

M. Sirsi.

Magnetoelectric Interaction Phenomena in Crystals. Edited by A. J. Freeman and H. Schmid. (Gordon and Breach Science Publishers), 1975. Price $ 19.50.

The book contains the invited lectures and the contributed seminar papers presented at the Symposium on Magnetoelectric Interaction Phenomena in crystals held at the Battelle Seattle Centre, Seattle, Washington, U.S.A., 21–24 May, 1973.

Magnetoelectric (ME) phenomena consist of the induction of a magnetization by an electric field, and the induction of an electric polarization by a magnetic field. The birth of this field was in 1956, when Landan and Lifshtiz showed from symmetry considerations that the phenomena could exist in their well known book on electrodynamics of continuous media (English Edition Addison–Wisley, 1960). Soon after, the effect was demonstrated in Cr_2O_3 by the work of Dzyaloshinskii, Astrov and Rado and coworkers. By 1961 both the electrically induced and the magnetically induced effects were demonstrated in Cr_2O_3.

In the search for additional ME materials, it was soon realized that only certain magnetic symmetry classes were compatible with the ME effect. A great majority of magnetically ordered crystals belong to the magnetic symmetry classes which forbid ME effects, and, hence it is not surprising that until 1970 only 13 ME materials were discovered. The growth of the field after 1970 has been very rapid and some practical applications appear to be possible in the near future.

The book under reviews has brought together the status of knowledge in this field upto 1973. Very useful introductory articles on various aspects of the subject are also included. For instance, Opechowski's article on magnetoelectric symmetry, Rado's survey of microscopic mechanisms of ME effects, O'Dell's survey of ME measurements, and Schmid's classification of ME Materials are excellent reviews, with sufficient introductory material. Other papers in the book cover recent investigations of most of the active workers is the field.

The book provides an excellent review for all workers in the field of magnetoelectric phenomena, crystal physics, magnetism, ferroelectricity, and structural phase transitions. I am sure the availability of this book will encourage many research workers to work in this area.

R. SRINIVASAN.

The Physical Principles of Electron Paramagnetic Resonance. By G. E. Pake and T. L. Estle. (Second Edition, W. A. Benjamin, Inc.), 1973. Price $ 19.50.

This is the second edition of the well known book by Professor G. E. Pake, published in 1962. The present edition too has been written for the advanced student and the research worker who have had a course in quantum mechanics. The subject of E. P. R. has become too vast to allow a single book to cover all aspects completely. This book therefore sets out to explain the basic principles of the subject and illustrates the applications to some problems in solid state physics.

Chapter II on the phenomenon of magnetic resonance is written with delightful clarity. The precision of spin systems is described with a number of figures in such a manner that it can be visualized by the reader. The next few chapters lay the foundation of the study of transition metal ions by EPR introducing the spin-Hamiltonian technique. A fairly complete treatment of the subject has been achieved. The chapter on interactions deals with the study of exchange in some detail and line shapes briefly.

Chapter VII describes the effect of stress and external electric fields in EPR. This topic has not been discussed in many books or reviews, and is thus, a particularly interesting and useful part of the book. The next two chapters on spin lattice relaxation and the effects of motion in liquids make an introduction to these two vast areas. The treatment of these topics is necessarily only brief and general. The examples in Chapter X are quite varied but are mainly from solid state physics. Double resonance is only briefly mentioned.

The book throughout contains references for further reading and this will be particularly appreciated by the beginner, attempting to enter this field in which the literature has already grown to such enormous proportions, in the total span of thirty years that the subject has been in existence. The text has been illustrated by a good number of figures.

The book is to be strongly recommended for advanced students of solid state physics, and to all research workers in magnetic resonance interested in problems in physics.

R. SRINIVASAN.

Elements of Electronic Navigation. By N. S. Nagaraja, Indian Institute of Science, Bangalore. (Publishers: Tata McGraw-Hill Publishing Company, Ltd., New Delhi-1), 1975. Pp. ix + 139. Rs. 15·00.

The book *Elements of Electronic Navigation* by N. S. Nagaraja is supposed to provide an introductory treatment of the subject to a student of Bachelor's degree in Electronics. The book describes the various physical concepts used in navigation in a very simple and lucid way. The author of the book seems to have definitely avoided going into a more quantitative treatment of the subject which, in my opinion, would have been more useful to a student of Bachelor's degree. However, the book does provide an overall view of the various navigational aids including the self-contained system of Doppler Radar and inertial navigation which should be quite useful to a beginner. The chapters on inertial guidance system and of recent developments such as Doppler VOR and Omega system are particularly well written.

The more serious drawback of the book is in its casual treatment of the recent developments in this field. Even if the subject of satellite navigation systems does not unfortunately form a part of the syllabus for the Bachelor's degree, I believe, the book would have been more useful if it had dealt with this subject in some greater detail to provide a Bachelor student at least a working concept of this subject. Particularly in veiw of the fact that a student of engineering does not have easy access to recent developments, whereas a large number of books are available on conventional navigational techniques, such an approach by the author would have had a great merit. Nevertheless, as an introductory book, it is well written and contains a number of references as well as exercises listed at the end of the book which should prove useful to a graduate student of communication engineering.

U. R. RAO.

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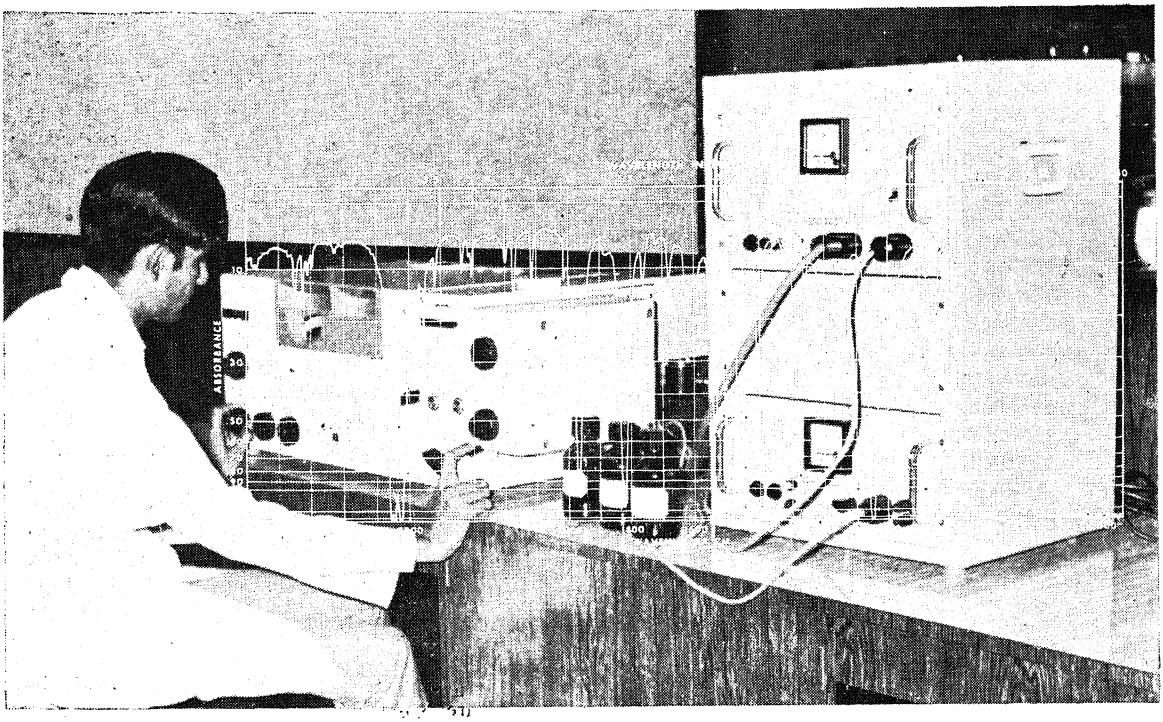

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$2^- \rightarrow 2^+$ BETA TRANSITION OF ^{42}K

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ABSTRACT

A Siegbahn-Slatis intermediate image magnetic coincidence spectrometer has been used to study the spectral shape of the non-unique first-forbidden beta transition in ^{42}K. The beta with an end-point energy of 2000 keV follows the spin sequence $2^- \rightarrow 2^+$ and has been studied in coincidence with the 1520 keV gamma that follows it. The shape has been fitted with a correction factor of the form $C(W) = k\,(1 + aW + b/W + cW^2)$ with $a = -0{\cdot}11 \pm 0{\cdot}060$, $b = 0{\cdot}701 \pm 0{\cdot}192$ and $c = 0{\cdot}012 \pm 0{\cdot}072$. The results of the present investigation are discussed from a suitable nuclear model interpretation point of view.

INTRODUCTION

THE decay scheme[1] of ^{42}K is well established. The outermost beta following the spin sequence $2^- \rightarrow 0^+$ with an end-point energy of 3520 keV is a first-forbidden unique transition[2]. The first inner beta component of present interest is a $2^- \rightarrow 2^+$ non-unique first-forbidden transition. Beta transitions with $\triangle J = 0$ like the $2^- \rightarrow 2^+$ transition in the case of ^{42}K are governed by all the six matrix element parameters. It is difficult to find unique solutions for these transitions in spite of accurate measurements of the beta decay observables. To add to this discrepancies in the measurements of various observables, persist because of the different experimental methods adopted by various authors. And as such measurements and re-measurements of the various beta decay observables governing the $2^- \rightarrow 2^+$ transitions are widely found in literature by the various groups of researchers using different instruments. This has prompted the authors to re-investigate the spectrum shape of the $2^- \rightarrow 2^+$ transition in ^{42}K with present set-up which yields accurate results.

The $2^- \rightarrow 2^+$ component of the present interest has a ξ-value $\left(\frac{aZ}{2R}\right) \approx 6{\cdot}8$ which is not much greater than the end-point energy W_0 ($\approx 4\ m_0 c^2$ units) thus suggesting the non-applicability of ξ-approximation to this case. There have been several investigations on this ($2^- \rightarrow 2^+$) inner beta of ^{42}K. Koertz[3] using a coincidence magnetic spectrometer and later Pohm *et al.*[4], using singles spectrometer independently reported an allowed shape in the energy region $0{\cdot}5$ to $1{\cdot}97$ MeV, within statistical fluctuations. This is in contradiction with high log *ft* value and the small value of ξ (compared to W_0). Andre *et al.*[5], using a Siegbahn–Slatis intermediate-image coincidence spectrometer and Daniel *et al.*[6], using a Heidelberg intermediate-image coincidence spectrometer report a considerable deviation from the allowed shape. They performed the shape factor analysis and fitted the data to a correction factor of the form $(C(W) = k\,(1 + aW + b/W + cW^2)$. The value of the constants *a*, *b* and *c* reported in the above two works differ from each other. It is, therefore, felt worthwhile to undertake a systematic investigation of this beta transition in ^{42}K using the present standadised set-up.

EXPERIMENTAL DETAILS

The radioactive material was produced by neutron irradiation of the enriched ^{41}K ($99{\cdot}49\%$) procured from ORNL, Tennessee, in the CIRUS reactor of BARC, Bombay. The radioactive material was obtained as a chloride in dilute HCl solution. The source was received in three consignments for the present experimentation. Uniform sources were prepared on thin aluminised mylar backing by vacuum evaporation. In general the source thicknesses for coincidence experiments ranged from 100–120 μgm/cm^2 and that of singles spectra from 30–60 μgm/cm^2. The sources were all of 2 mm diameter. The total time taken for each run was restricted to 8 hours to reduce the decay correction.

A Siegbahn–Slatis β-ray spectrometer is used to investigate the beta spectrum. The optimisation of the spectrometer for precision analysis of beta spectra and elimination of back scattering effects, etc., have been described elsewhere[7-9]. The details of the modifications, optimisation of the spectrometer for coincidence studies and its operation have been described elsewhere[10-12].

An overall test of the equipment was obtained from a measurement of the coincidence spectrum of the 960 keV β group in ^{198}Au, under conditions similar to those used in the ^{42}K experiment. The shape factor plot was fitted to a linear correction factor of the form $C(W) = k\,(1 + aW)$ with $a = (-\ 0{\cdot}025 \pm 0{\cdot}008)\ (m_0\, c^2)^{-1}$ and the end-point energy of the beta is 962 ± 2 keV. This result is found to be in excellent agreement with the earlier reported works[13-16].

The coincidence spectrum was recorded by scanning the beta spectrum in coincidence with the 1520 keV gamma rays. The resolving time of the coincidence unit was set at 24 ns. The 1·52 MeV gammas were accepted in a window of 30% of the photopeak in the gamma channel. All spectra were roughly scanned in steps of 50 keV in the energy range 200 to 1850 keV. For each run about 3,000 counts were collected at the maximum of the beta continuum. The observed count rate was corrected for (*i*) chance rate, (*ii*) finite spectrometer resolution, (*iii*) back-scattering, (*iv*) coincidence efficiency and (*v*) beta-gamma angular correlation anisotropy.

The shape of this transition was also analysed from the singles experiment. The singles spectrum was recorded in the range 0·200 to 3·5 MeV in steps of 60 keV. The outer beta with a spin change ($\triangle$J) equal to 2 is unique firstforbidden transition and has been analysed[2] fitting the spectrum to a correction factor for the form

$$C(U) = (q^2 + 9\,L_1/L_0)\;(1 - a\,W).$$

The shape of the inner beta component was constructed by eliminating the influence of the outer beta by the subtraction method.

The spectra were analysed by standard procedures described earlier[7-9]. Figure 1 shows the F.K. plot of the coincidence spectrum. The correct end-point energy was judged from the behaviour of the shape factor curves when W_0, the end-point energy, is varied in steps of 2 keV. The experimental shape factor was weighted least-square fitted to a shape correction factor of the form $C(W) = k(1 + aW + b/W + cW^2)$ and is represented by the dashed line in Fig. 2, for the data corresponding to the run No. 2. The results due to the three coincidence runs and the singles run are summarised in Table I. From Table II, it can be seen that the present results concerning the values of end-point energy are in substantial agreement with those of Andre[5].

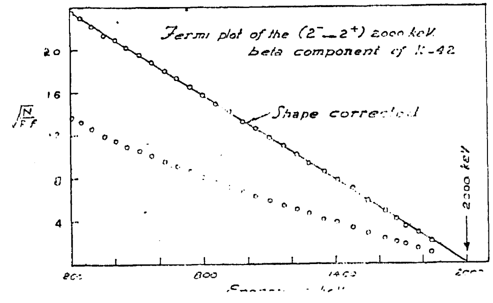

FIG. 1. Fermi plot of K-42 bata spectrum in coincidence with the 1520 keV gamma-ray.

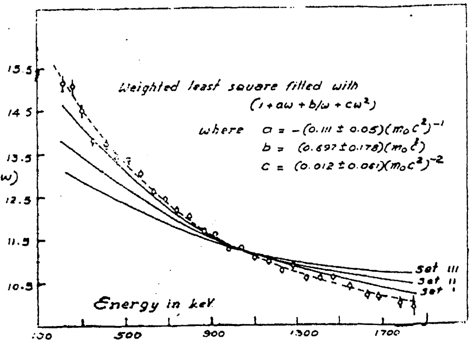

FIG. 2. Shape-factor of ^{42}K (2000 keV). The experimental points are fitted with $c(W) = k(1 + aW + b/W + cW^2)$ by weighted least squares. The three solid lines represent the theoretical estimates from the matrix elements sets due to Vema Reddy *et al.*

DISCUSSION

First-forbidden transitions with no spin change are in general too involved to facilitate a matrix element analysis as these transitions are caused by as many as six matrix elements. However, it is interesting to note that most of these transitions exhibit the characteristics of an allowed beta transition and can be well described by the ξ-approximation or the so-called 'quasi-allowed approximation'. This is not surprising as a cancellation effect is not likely to take place simultaneously in both V and Y. As can be seen from shell model, a reduction in Y by an accidental cancellation is also highly improbable, except in the case where Λ_1, in equation $Y = -C_V\,\xi\,(\Lambda_{\text{cvc}} - 1 - 1{\cdot}2\,\Lambda_1) \int i\vec{r}$, is about 1. The large spectrum shape deviation (of the order of 35%) as found in the present work and also the large beta-gamma anisotropy concerning the $2^- \rightarrow 2^+$ beta transition in ^{42}K besides a high log *ft* value show the breakdown of ξ-approximation in this case. Also, the condition for ξ-approximation, namely, $\xi \gg (W_0 - 1)$ is not satisfied inasmuch as that the ratio of $(W_0 - 1)/\xi \approx 0{\cdot}6$ for this transition. Thus, this transition can only be classified as one that deviates from ξ-approximation[17] and this deviation may perhaps be due to an accidental cancellation in Y or V or due to a selection rule effect. Vema Reddy *et al.*[18] have evaluated, using Simms[19] accurate formalism, the six matrix element parameters associated with this transition in ^{42}K with their beta-gamma angular correlation results coupled to the shape factor due to the present

TABLE I

Results of the beta shape factor analysis of the $2^- \rightarrow 2^+$ *beta in* ^{42}K

Run No.		E_0 (keV)	Shape factor $C(W) = k(1 + aW + b/W + cW^2)$		
			$a\,(m_0c^2)^{-1}$	$b\,(m_0c^2)$	$c\,(m_0c^2)^{-2}$
Coincidence	1	2001±2	−0·111±0·06	0·701±0·192	0·012±0·072
	2	2000±2	−0·111±0·05	0·697±0·178	0·012±0·061
	3	2003±2	−0·114±0·05	0·662±0·182	0·012±0·063
Singles	1	1995±2	−0·324±0·023	1·270±0·088	0·035±0·002

TABLE II

Comparison of the end-point energy and the shape correction factor constants due to different authors for the 2 MeV transition of ^{42}K

Author	E_0 (keV)	$C(W) = k(1 + aW + b/W + cW^2)$		
		$a\,(m_0c^2)^{-1}$	$b\,(m_0c^2)$	$c\,(m_0c^2)^{-2}$
Pohm[4]	1980	$0 \leq a \leq 0\cdot1$	..	..
Andre[5]	2000	−0·12±0·04	0·67±0·06	0·013±0·008
Daniel[6]	2000	−0·15±0·026	0·81±0·47	−0·020±0·030
Present work	2000	−0·11±0·05	0·70±0·178	0·012±0·061

TABLE III

Matrix element parameter sets reported by Vema Reddy et al.[6], *consistent with the present shape measurements*

	Y	V	x	u	w	z	λ	Λ_{cvc}
Set I	0·440	2·28	−2·56	−4·28	−2·70	1·00	0·25	3·10
Set II	0·500	2·28	−2·56	−4·18	−2·70	1·00	0·32	3·03
Set III	0·378	2·20	−2·35	−3·82	−1·60	1·00	0·29	3·06

investigation and the circular polarisation results due to De Saitignon[20]. Figure 2 shows the shape predicted by three sets due to this analysis and the corresponding values are tabulated in Table III. From Fig. 2 it can be seen that the shape predicted by Set 1 is in good agreement with the present measurement. On examining the various values of the parameters, especially the value of Y which is reduced by one order of magnitude when compared to other matrix element parameters one can only conclude that there is a significant suppression in the magnitude of Y which could be due to an accidental cancellation and this is in clear agreement with the conclusion we arrived at earlier when we considered the applicability of ξ-approximation to the present behaviour of the beta spectral shape.

Applicability of Nuclear Models to the $2^- \rightarrow 2^+$ *Transition.*—^{42}K is an odd-odd nucleus with 19 protons and 23 neutrons decaying *via* beta decay to the even-even daughter nucleus ^{42}Ca, with 20 protons and 22 neutrons. In the shell description the parent state can be given as $1\,f_{7/2}$ while the daughter state as $1\,d_{3/2}$, a neutron transforming into a proton. In a transition of this kind where $\triangle J = 2$, only the rank 2 matrix element $\int B_{ij}$ should be different from zero and the shape exhibited by such a transition should be of the unique first-forbidden type. But, this is not true in the present case, when one examines the shape factor plot of the beta spectrum and also the values of the nuclear matrix element parameters due to the analysis of Vema Reddy *et al.*[18] inasmuch as the rank zero and one matrix elements make significant contributions to this transition. From this it may be concluded that the 2$^-$ and 2$^+$ states of ^{42}K and ^{42}Ca respectively cannot be represented as pure shell model states. Andre *et al.*[5], suggested an admixture of $2p_{3/2}$ in the parent state. Then, the transition may follow the spin sequence $2p_{3/2} \rightarrow 1d_{3/2}$. In this case where $\Delta J = 0$, all the six matrix element belonging to rank 0, 1 and 2 govern the beta transition. Following this an attempt is made to compute the matrix element ratios x/z, u/z and w/z employing the expressions of Rose and Osborn[21]. The values thus obtained are given below.

$$x/z = -0\cdot2976,\ u/z = -1.4140 \text{ and}$$
$$w/z = -1\cdot3691.$$

A comparison of these values with the experimentally derived ones given in Table III, shows that even this description based on an admixture of configurations does not give a correct picturisation of the involved parent state. Hess[22] suggests another alternative to explain these matrix element parameters by introducing a deformed component both in the parent and the daughter states. This requires, the vanishing of rank one matrix elements. From the present solutions of matrix element parameters, it can be seen that these matrix elements are different from zero. Thus the suggestion of Hess also appears to be not true for the present case. From a model point of view both the parent and the daughter states are not well understood. Finally one can only conclude that a theoretical investigation of the present involved state from several angles would be highly useful in explaining the present trend of this 2·0 MeV transition in ^{42}K.

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1. *Nuclear Data Sheets*, 1974, A214.
2. Narasimha Rao, C., Mallikarjuna Rao, B., Mallikharjuna Rao, P. and Venkata Reddy, K., *Phys. Rev. C*, April, 1975, **11**, 735.
3. Koertz, Schwarchild, Gold, and Wu, *Phys. Rev.*, 1954, **95**, 612.
4. Pohm, A. V., Waddell, R. C. and Jenson, E. N., *Ibid.*, 1956, **101**, 1315.
5. Andre, S. and Depommier, P., *Compt. Rend.*, 1964, **259**, 1102.
6. Daniel, H., Schmitt, H. and Springer, K., *Kolloquim Uber Betazerfall u Schwache Wechselwirkung*, Max Plant Institut fur Kern–Physik, Heidelberg, 1965, p. 225.
7. Nagarajan, T. and Venkata Reddy, K., *Nucl. Inst. Meth.*, 1969, **66**, 77.
8. — and —, *Phys. Rev.*, 1969, **178**, 1968.
9. — and —, *Nucl. Instr. Meth.*, 1970, **80**, 217.
10. Mallikarjuna Rao, B., *Ph.D. Thesis*, Andhra University, Waltair, 1975 (Unpublished).
11. —, Narasimha Rao, C., Mallikharjuna Rao, P. and Venkata Reddy, K., Communicated to *Nucl. Instr. Meth.*
12. Ravindranath, M., Narasimha Rao, C. and Venkata Reddy, K., *Nucl. Phys. and Solid State Phys. Symp.*, Bombay, 1972, **14 B**, 511.
13. Keeler, W. J. and Connor, R. D., *Nucl. Phys.*, 1965, **61**, 513.
14. Lewin, W. H., Van Nooyen, B., Van Eijk, C. W. E. and Wapstra, A. H., *Ibid.*, 1963, **48**, 159.
15. Paul, H., *Ibid.*, 1965, **72**, 326.
16. Van Krugten, H., *Thesis*, Univ. of Tech. Delft, 1967 (Unpublished).
17. Kotani, T. and Ross, M., *Phys. Rev. Lett.*, 1958, **1** (4), 140.
18. Vema Reddy, B., *Ph.D. Thesis*, Andhra University, Waltair, 1974.
19. Simms, P. C., *Phys. Rev.*, 1965, **138 B**, 784.
20. Saitignon, P. De., Chabre, M. and Depommier, P., *Compt. Rend.*, 1964, **258**, 884.
21. Rose, M. E. and Osborn, R. S., *Phys. Rev.*, 1954, **93**, 1326.
22. Hess, R., *Helv. Phys. Acta*, 1968, **41**, 77.

SYNTHESIS OF GLABRANINE, 5, 7-DIHYDROXY-6-PRENYL-FLAVANONE AND 5-HYDROXY-7-METHOXY-6-PRENYL FLAVANONE

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ABSTRACT

An unambiguous synthesis of glabranine (I), 5, 7-dihydroxy-6-prenyl flavanone (II) and 5-hydroxy-7-methoxy-6-prenyl flavanone (III) have been carried out starting from phloroglucinol and cinnamoyl chloride. Syntheses of two chromeno-flavanones have also been reported.

GLABRANINE (I) m.p. 154-55° was isolated from *Glycyrrhiza glabra*[1] and has been assigned the structure. 8-C-prenyl-5, 7-dihydroxy-flavanone from its spectral studies. Recently an isomer of Glabranine, 5, 7-dihydroxy-6-prenyl flavanone (II) and the methyl ether (III) have also been isolated from Derris Rariflora[2] and the structures established based on spectral data. They have now been synthesised and the properties of the synthetic compounds have been found to be in complete agreement with those reported in the literature.

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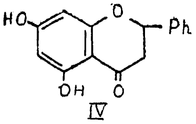
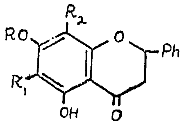
Treatment of phloroglucinol with cinnamoyl chloride and aluminium trichloride in nitrobenzene yielded 5, 7-dihydroxy flavanone[3] (IV), m.p. 170–75°. The compound (IV) (1 g) on treatment with 2-methyl-but-3-en-2-ol (0·4 ml) and boron trifluoride-etherate in dry dioxan for one hour at room temperature gave a mixture of five compounds, as shown by TLC, one of which was the starting compound. The reaction product was taken up in ether and the ether solution extracted with 1% sodium carbonate and 1% sodium hydroxide respectively. The sodium carbonate soluble fraction yielded the unreacted starting material (60% recovered), The sodium hydroxide soluble fraction was found to be a mixture of two components and was subjected to column chromatography over silica gel.

Elution with petroleum ether-benzene (7:3), yielded a colourless compound (II) (40 mg), crystallised from petroleum ether-benzene, ($C_{20}H_{20}O_4$), m.p. 214–16°, λ_{max} 295 nm, ν_{max}^{Nujol} 3350, 1650, 1625, 1575 cm^{-1}. It gave dark brown colour with ferric chloride and agreed with 5, 7-dihydroxy-6-prenyl-flavanone in all respects[3] (lit. m.p. 212–14°). The compound was methylated with 1·1 mole of dimethyl sulphate in acetone containing potassium carbonate. The compound III crystallised from methanol as colourless needles, ($C_{21}H_{22}O_4$), m.p. 98–99° (lit. m.p. 90–92°), ν_{max}^{Nujol} 3375, 1640, 1617 cm^{-1}. It gave brown colour with ferric chloride. The NMR ($CDCl_3$) of the methyl ether showed the following peaks: δ 12·03 (*s*, 1H, chelated $-OH$), 7·26 (*s*, 5H, $-C_6H_5$), 5·88 (*s*, 1H, C_8-H), 4·86–5·36 (*m*, 2H, C_2-H and $> C = CH-$), 3·72 (*s*, 3H, OCH_3), 3·10 (*d*, J = 7 Hz, 2H, $Ar-CH_2-CH = C <$), 2·60–2·90 (*m*, 2H, C_3-H_2) 1·60 and 1·70 $\left((2s, 6H, = C\begin{array}{l}CH_3\\ CH_3\end{array}\right)$.

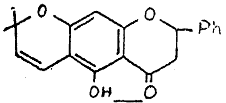
Elution of the column with petroleum ether-benzene (1:1) yielded the other compound I, colourless needles (50 mg) from petroleum ether-benzene, ($C_{20}H_{20}O_4$), m.p. 155–57° giving dark brown colour with ferric chloride; λ_{max} 297 nm; ν_{max}^{Nujol} 3360, 1645, 1620, 1560, 1520 cm^{-1}. NMR ($CDCl_3$): δ 7·28 (*s*, 5H, $-C_6H_5$), 5·91 (*s*, 1H, C_6–H), 4·90–5·50 (*m*, 2H, C_2-H and $> C = CH-$), 3·17–3·47 (*m*, 2H, $Ar-CH_2-CH = C<$), 2·75–3·00 (*m*, 2H, C_3-H_2), 1·66 and 1·68 $\left(2s, 6H, = C\begin{array}{l}CH_3\\ CH_3\end{array}\right)$ The properties of the compound were found to be in complete agreement with those reported for glabranine[1] (lit. m.p. 154–55°). Compound I was methylated using 1·1 mole of dimethyl sulphate and potassium carbonate in acetone and the methyl ether (V) crystallised from methanol, ($C_{21}H_{22}O_4$), m.p. 110–11°; ν_{max}^{KBr} 3375, 1640, 1620 cm^{-1}; NMR (CCl_4): δ 11·95 (*s*, 1H, chelated $-OH$), 7·2 (*s*, 5H, $-C_6H_5$), 5·90 (*s*, 1H, C_6-H), 4·80–5·40 (*m*, 2H, C_2-H and $> C = CH-$), 3·75 (*s*, 3H, $-OCH_3$), 3·10 (*d*, J = 7 Hz, 2H, $Ar-CH_2-CH = C <$), 2·70–2·90 (*m*, 2H, C_3-H_2), 1·58 $\left(s, 6H, = C\begin{array}{l}CH_3\\ CH_3\end{array}\right)$. To confirm that glabranine (I) is the 8-prenyl flavanone its 7-0-methyl ether (V) was subjected to treatment with DDQ in boiling xylene for 16 hours when V was recovered unchanged. This proved that the prenyl group in V and I was at 8 and not at 6 position. Had it been at 6, it would have resulted in a cyclised product. Treatment of 7-0-methyl ether (III) of II under the same conditions resulted in a cyclised product devoid of ferric reaction, proving thereby that II is the 6-prenyl flavanone. The cyclised product was very small to do further investigations.



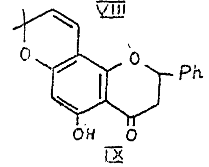
IV



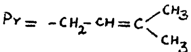
I $R = R_1 = H$, $R_2 = Py$
II $R = R_2 = H$, $R_1 = Py$
III $R = CH_3$, $R_1 = Py$, $R_2 = H$
V $R = CH_3$, $R_1 = H$, $R_2 = Py$
VI $R = Py$, $R_1 = R_2 = H$
VII $R = H$, $R_1 = R_2 = Py$



VIII



IX



$Py = -CH_2-CH=C\begin{array}{l}CH_3\\ CH_3\end{array}$.

The remaining ether solution was found to be a mixture of two compounds and was subjected to column chromatography over silica gel. On elution with petroleum ether a colourless semi-solid (VI) (40 mg) was obtained which could not be crystallised but was single on TLC (Benzene). It gave brown colour with ferric chloride. λ_{max} 285 nm; ν_{max}^{Nujol} 3360, 1645, 1575, 1565 cm^{-1}; NMR (CCl_4): δ 7·2 (*s*, 5H, $-C_6H_5$), 5·78 (*s*, 2H, C_6–H and C_8–H), 4·95–5·40 (*m*, 2H, C_2–H and $> C = CH-$), 4·32 (*d*, J = 7 Hz, 2H, $-O-CH_2-CH=$), 2·50–2·85 (*m*, 2H, C_3-H_2), 1·61, 1·68 $\left(2s, 6H, = C\begin{array}{l}CH_3\\ CH_3\end{array}\right)$. The NMR showed that the compound VI was 7-0-prenyl-5-hydroxy-flavanone and this was confirmed by 0-prenylation of IV using prenyl bromide in acetone containing potassium carbonate when the semi-solid obtained was found to be identical with VI (Co-TLC and I.R.). Further elution of the column with petroleum ether-benzene (1:1) yielded a solid which crystallised from petroleum ether-benzene as colourless needles (30 mg), ($C_{25}H_{28}O_4$) m.p. 100–101°; brown colour with ferric chloride; ν_{max}^{Nujol} 3360, 1625, 1605 cm^{-1}. The NMR of the compound showed it to be 5, 7-dihydroxy-6, 8-di-C-prenyl flavanone (VII).

Since a number of hydroxy chromenoflavanone are known to occur in Nature, it was considered worthwhile to prepare the two isomeric chromenes from II and I. Thus, when II was treated with DDQ in benzene solution for two hours the corresponding chromene (VIII) was obtained in 70% yield, as colourless needles from methanol, ($C_{20}H_{18}O_4$), m.p. 132–33°; brown colour with ferric chloride; λ_{max} 270, 292 nm; ν_{max}^{Nujol} 1650 cm^{-1}. NMR ($CDCl_3$): δ 12·46 (*s*, 1H, chelated -O*H*), 7·45 (*s*, 5H, C_6H_5), 6·54 (*d*, J = 11 Hz, 1H, Ar-C*H*-CH-Č-O), 6·01 (*s*, 1H, C_6–*H*), 5·26–5·49 (*m*, C_2–*H* and Ar–CH = C*H*–Č–O–), 2·73–3·03 (*m*, 2H, C_3–H_2), 1·44 $\left(s, 6H, > C\begin{smallmatrix}CH_3\\CH_3\end{smallmatrix}\right)$.

A similar reaction with I yielded the chromene (IX) in 70% yield. It was crystallised as colourless needles from methanol, ($C_{20}H_{18}O_4$), m.p. 100–101°; brown colour with ferric chloride; λ_{max} 266, 292 nm; ν_{max}^{Nujol} 1650 cm^{-1} NMR ($CDCl_3$): δ 12·41 (*s*, 1H, chelated -O*H*), 7·45 (*s*, 5H, –C_6H_5), 6·64 (*d*, J = 11 Hz, Ar–C*H* = CH–Č–O–), 6·00 (*s*, 1H, C_8–*H*), 5·25–5·61 (*m*, 2H, C_2–*H*) and Ar–CH = C*H*–Č–O–), 2·80–3·11 (*m*, 2H, C_3–H_2), 1·44 $\left(s, 6H, > C\begin{smallmatrix}CH_3\\CH_3\end{smallmatrix}\right)$.

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1. Kattaev, N. Sh. and Nikonov, G. K., *Khim. Prir. Soedin.*, 1972, **6**, 805 (*C.A.*, **78**, 84806*c*).
2. Filhot, R. B., Gottlieb, O. R. and Mourao, A. P., *Phytochemistry*, 1975, **14**, 261.
3. Tatuta, Haruo, *J. Chem. Soc. Japan*, 1940, **61**, 752, (*C.A.*, **37**, 376^6).

DECRYPTIFICATION OF NITRATE REDUCTASE ACTIVITY IN THE YEAST *CANDIDA UTILIS*

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ABSTRACT

A diverse variety of chemicals and drugs are known to affect the cell walls of bacteria and yeasts resulting in an enhanced access of the intracellular enzymes to the added substrates. The ability of organic solvents, antifungal drugs, detergents, and chelating compounds to decryptify NAD(P)H: nitrate oxidoreductase (EC 1.6.6.2) activity in *Candida utilis*, has been investigated. Of these, toluene, butanol, 2-phenylethanol and amphotericin B, in that order, are effective. The degree of decryptification is dependent on the concentration of the agent, and at 2·5% (v/v), toluene-ethanol elicited highest enzyme activity. These permeated cell preparations greatly aid in the regulatory studies of various yeast enzymes *in situ*.

INTRODUCTION

INVESTIGATIONS concerning regulatory phenomena, *viz.*, induction and repression of various enzymes such as β-galactosidase in *Escherichia coli* were carried out *in situ* using permeated bacterial cells[1-3], thus doing away with the laborious process of preparing cell-free extracts. The growing awareness in the recent past of the fallacies inherent in inferring the *in vivo* situation from *in vitro* observations further strengthened the surge of interest in the development of novel methods to decryptify various enzymes and assay the activities *in situ* in permeated cells of a variety of organisms[4-10].

The detailed studies on the regulation of yeast nitrate reductase (EC 1.6.6.2) (NAR) under investigation in our laboratory which involve determination of rapidly changing enzyme levels[8], necessitated the development of an alternate assay method, which is simpler, rapid and more sensitive than the conventional *in vitro* procedure.

The paper is concerned with the examination of the relative efficacy of different compounds in decryptifying NAR activity in induced *Candida utilis* cells. The typical method used to assay the decryptified NAR activity *in situ* using permeated *C. utilis* cells is also described.

MATERIALS AND METHODS

Chemicals.—Lab reagent grade benzene was purchased from Sarabhai M. Chemicals, and solvent ether from Alembic Chemical Works Co. Ltd., Baroda. Analytical reagent grade chloroform, DMSO, DMF, acetone, propanol, butanol, amyl alcohol, potassium nitrate, sulfur-free grade lab reagent toluene and polyethylene glycol 400 were purchased from BDH Ltd., Bombay, India. 2-Phenylethanol, amphotericin B, nystatin, griseofulvin, sodium salt of EDTA, NADH, FAD, DTT, and crystalline bovine serum albumin were obtained from Sigma Chemical Co., St. Louis, U.S.A. Miconazole (1-{2-(2, 4-dichlorophenyl)-2-[(2, 4-dichlorophenyl) methoxy]ethyl}-1 H-imidazole mononitrate) was a kind gift from

Janssen Pharmaceutica, B-2340, Beerse, Belgium, through the courtesy of Ethnor, Ltd., Bombay. Sulfanilamide was purchased from the Indian Drugs and Pharmaceuticals, Ltd., India and N-(1-naphthyl) ethylenediamine. 2 HCl from BDH, Ltd., Poole, England. Other chemicals were of analytical grade from BDH Ltd., Bombay, India or Riedel–De Haenag Seelze–Hannover, Germany.

Organism and Maintenance.—Candida utilis CBS 4511 was obtained from the Centraalbureau voor Schimmelcultures, Delft, The Netherlands. The yeast was maintained by subculturing once in two months on Sabouraud's glucose agar slants containing neopeptone, 1% ; glucose, 1% and yeast extract, 0·2%.

Media and Cultivation Techniques.—The basal medium was essentially the same as described by Wickerham[11] with a few modifications, and contained potassium nitrate, 50 mM (N) as nitrogen source (induction medium). The yeast was cultured by inoculating liquid medium in 500 ml-Erlenmeyer flasks with 5% (v/v) of 24 h culture, aerated by shaking on an Emenvee gyratory shaker (180 rpm) at 30° C. The cells in late exponential phase of growth ($\simeq$ 12 h after inoculation) were harvested, washed with chilled phosphate buffer, and packed by centrifugation at 6,000 × g for 5 min. at 4° C in a Sorvall RC 2–B centrifuge. The packed cells were stored frozen at $-$ 10° C till use.

Induction of NAR.—The cells of *C. utilis* grown on KNO_3 (1%, w/v) as nitrogen source had high levels of NAR activity. The inducibility of NAR in *C. utilis* by NO_3^- was reported[12].

Preparation of Permeated Yeast Cells.—The general procedure of permeating the yeast cells was as follows: An aliquot of 0·05 ml chilled decryptifying agent was added to 2 ml of the cell suspension [100 mg (fresh wt) of induced cells/ml of 'preparation buffer', 0·1 M phosphate buffer, pH 7·0 containing 10^{-4} M DTT and $1{\cdot}5 \times 10^{-4}$ M EDTA] in a 16 mm test-tube, and shaken for 2 min at 4° C on a Jay Vortex Mixer. Suitable aliquots of this preparation were used to assay NAR *in situ*.

Enzyme Assay :

NAR in situ.—The reaction mixture contained, KNO_3, 0·02 M ; 0·5 ml (50 mg fresh wt) of permeated cell preparation, and 0·1 M pyrophosphate buffer, pH 7·0 to give a final volume of 1 ml. The reaction was carried out 10 min. at 37° C and terminated by the addition of 1 ml of 1% sulfanilamide in 3 N HCl. The nitrite content of an aliquot of the supernatant obtained after low speed centrifugation was determined. There was no significant reduction of nitrite in blanks under the assay conditions.

Analytical Assay :

Nitrite.—The nitrite content of 1 ml-reaction mixture was determined colorimetrically by a modification of the diazo coupling procedure described by Snell and Snell[13], with 1 ml of 1% (w/v) sulfanilamide in 3 N HCl followed by 1 ml of 0·02% (w/v) N-(1-naphthyl) ethylenediamine dihydrochloride, and distilled water to give a final volume of 5 ml. After 10 min, the color intensity was determined at 540 nm with a Beckman model DU spectrophotometer. The absorbancy values were converted into nmoles of nitrite by multiplication with the slope of a nitrite concentration reference curve made with sodium nitrite.

Units and Specific Activity.—One unit of activity is defined as equivalent to that amount of the enzyme which catalyzes the formation of 1 nmole of nitrite per 1 min and the specific activity is expressed as units per mg of total extractable protein (55 mg/g fresh yeast).

RESULTS

Relative Efficacy of Various Permeating Agents in Decryptifying NAR Activity in C. utilis.—To select a suitable permeating agent that can effectively decryptify NAR in *C. utilis,* cells from exponential phase of growth were harvested, washed, and suspended in the preparation buffer to give 100 mg (wet wt) of cells/ml, and treated with various permeating agents reported in the literature, such as different organic solvents including alcohols, membrane-affecting drugs, chelating agents, etc., according to the typical procedure, described under Materials and Methods. A comparison of the relative levels of NAR activity of the cells permeated by different solvents showed toluene to be the most effective, and benzene the next best (Table I). Acetone, DMSO, chloroform, ether and DMF were not efficient as permeating agents.

Butanol, followed by ethanol, was the best among the aliphatic alcohols tested in decryptifying NAR activity. Phenylethanol, the only aromatic alcohol tested was almost as effective as butanol. Although polyethylene glycol 400 and propanol did show some effect, the relative levels of NAR activity decryptified, however, were not significant.

A number of antifungal drugs, known to affect cell membranes of the susceptible organisms, were also tested for their efficacy in permeating yeast cells. Amphotericin B was most effective folowed by miconazole, a synthetic compound recently coming into increasingly wide use as a broad-spectrum antimicrobial drug,

TABLE I

Decryptification of nitrate reducatase activity in C. utilis *by various compounds*

Solvent[a]	NAR[c] specific activity
Organic solvents:	
Toluene	18·0
Benzene	9·9
Acetone	1·1
DMSO	0·1
Alcohols:	
Butan–1–01	16·2
2-phenylethanol	14·7
Ethanol	13·2
Polyethylene glycol 400	7·8
Propan–2–ol	5·4
Membrane-active drugs[b]:	
Amphotericin B	12·0
Miconazole	8·0

a used at a concentration of 5% (V/V).
b The final concentration was 1 mM.
c Assayed *in situ* at 37°C for 10 min.

Nystatin and griseofulvin, at the concentrations tested, had little effect. Various detergents like Triton X-100, Brij, sodium deoxycholate either failed to bring about any decryptification of NAR activity or inactivated the enzyme. Similarly, EDTA[5] and bovine serum albumin[7] used by earlier workers as permeating agents, were not useful in decryptifying NAR activity of *C. utilis* cells.

Effect of Concentration of the Permeating Agent on Decryptification of NAR Activity.—Further, the effect of varying the concentration and composition of the permeating agent on decryptification of NAR activity was investigated. Figure 1 shows the effect of different concentrations of toluene, ethanol and their mixture on permeation of the yeast cells. Although toluene was very effective in permeating the yeast cells, its effect was enhanced significantly, when it was used in combination with ethanol, as indicated by increased levels of NAR activity. Maximal enzyme activity was elicited by 2·5% (v/v) toluene-ethanol (1 : 4, v/v). Higher concentrations of toluene-ethanol, however, were deleterious to the enzyme activity[14]. This could be due to the probable sensitivity of the enzyme to alcohol because toluene, by itself did not affect NAR activity at higher concentrations. Untreated cells, however, showed no evidence of decryptified NAR activity,

DISCUSSION

Of various permeating agents examined, toluene, in combination with alcohol, was most effective in decryptifying NAR activity in *C. utilis* cells. A variety of substances including toluene[1–3,16,17] benzene[6], DMSO[8], polyene antibiotics[4], basic proteins[7], and chelating compounds[5], was reported to permeate bacterial and yeast cells. Toluene found extensive use in such studies for its selective action on the cells rendering them freely permeable to a wide variety of low molecular weight substances[16,17]. Our results substantiate this observation regarding the high retention of the enzymatic potential of the treated cells. The absence of NAR activity in cells treated with EDTA (widely used by other workers to permeate cells) may be the result of the inhibition by EDTA of the activity of NAR, a metalloflavo-protein rather than its failure to permeate yeast cells[14].

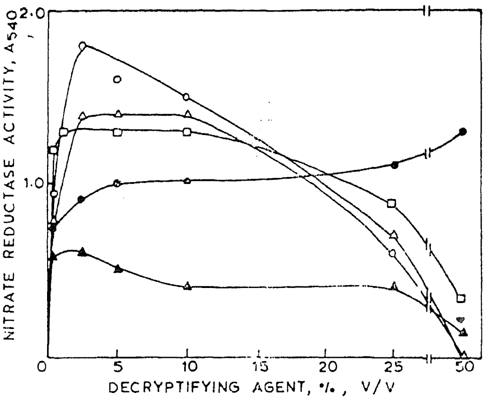

FIG. 1. Decryptification of nitrate reductase activity in *C. utilis* as a function of the concentration of the permeating agent. An O.D. of 1·0 at 540 nm corresponds to 10 nmoles of nitrite formed/min/mg protein. (●) toluene; (▲) ethanol; (□) toluene-ethanol. 1 : 1, v/v; (△) toluene-ethanol, 1 : 2, v/v; (○) toluene-ethanol, 1 : 4, v/v.

The toluene-treated cells showed high NAR activity under *in situ* conditions without added cofactors[10], contrary to the property of NAR *in vitro*, where the enzyme showed an absolute requirement for NAD (P) H[14,15]. However, the addition of coenzymes increased the *in situ* activity by 50%. The initial high *in situ* activity could be due to the participation of endogenous NAD (P) H and FAD in the reaction. Presence of adequate intracellular concentrations of NADH and cAMP in yeast cells was recently reported by Gancedo and Gancedo[18].

The proposed *in situ* assay, thus, is much simpler, less expensive, quite sensitive and reliable, and enables one to overcome the apparent shortcomings attributed to the *in vitro* method of assay. The additional advantage that the cells need no other supplement of the coenzymes for routine assay of the enzyme activity *in situ,* makes this procedure uniquely distinct even over the other *in situ* assay methods known, where the only advantage over the conventional *in vitro* methods is that whole cells can be used in place of cell-free preparations. All our subsequent studies on the regulatory phenomena controlling the biosynthesis of NAR in *C. utilis,* which involve handling of innumerable concentrations, and assay of NAR activities at brief time periods, are conducted using the *in situ* technique and the results appear elsewhere.

ACKNOWLEDGEMENT

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1. Pardee, A. B., Jacob, F. and Monod, J., *J. Mol. Biol.,* 1959, **1**, 165.
2. Kempfer, R. O. R. and Magasanik, B., *Ibid.,* 1967, **27**, 475.
3. Reeves, R. E. and Sols, A., *Biochem. Biophys. Res. Commun.,* 1973, **50**, 459.
4. Cirillo, V. P., Harsch, M. and Lampen, J. O., *J. Gen. Microbiol.,* 1964, **35**, 249.
5. Leive, L., *Proc. Natl. Acad. Sci. U.S.A.,* 1965, **53**, 745.
6. Magee, P. T. and de Robichon-Szulmajster, H., *Eur. J. Biochem..* 1968, **3**, 507.
7. Schlenk, F. and Zydek-Cwick, C. R., *Arch. Biochem. Biophys.,* 1970, **138**, 220.
8. Adams, B. G., *Anal. Biochem.,* 1972, **45**, 137.
9. Jaworski, E. G., *Biochem. Biophys. Res. Commun.,* 1971, **43**, 1274.
10. Radin, J. W., *Plant Physiol.,* 1973, **51**, 332.
11. Wickerham, L. J., *J. Bacteriol.,* 1946, **52**, 293.
12. Choudary, V. P. and Ramananda Rao, G., *Abstr. 14th Ann. Conf. Assoc. Microbiol. India,* 1973, **33**, 2.
13. Snell, F. D. and Snell, C., *Colorimetric Methods of Analysis,* Van Nostrand Co., New York, 1949, **2**, 804.
14. Choudary, V. P., *Ph.D. Thesis,* Indian Institute of Science, Bangalore, India, 1975, p. 57.
15. — and Ramananda Rao, G., *Proc. Ind. Acad. Sci.,* 1976 (In press).
16. Jackson, R. W. and DeMoss, J. A., *J. Bacteriol.,* 1965, **90**, 1420.
17. Peterson, R. L., Radcliffe, C. W. and Pace, N. R., *Ibid.,* 1971, **107**, 585.
18. Gancedo, J. M. and Gancedo, C., *Biochimie,* 1973, **55**, 205.
19. Choudary, V. P. and Ramananda Rao, G., *Can. J. Microbiol.,* 1976, **22**, 35.

A RAPID SPECTROPHOTOMETRIC METHOD FOR THE QUANTITATIVE DETERMINATION OF DIOSGENIN IN *DIOSCOREA* TUBERS

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ABSTRACT

A rapid method for the assay of diosgenin in *Dioscorea* tubers has been developed. Simultaneous hydrolysis and extraction of the tubers (50 mg) by 3 N HCl and hexane is carried out in 1½ to 2 hours. $SbCl_5$ (24% in $HClO_4$) has been used for developing colour. The red colour formed is stable for 2 hours and is evaluated spectrophotometrically at 486 nm. One analyst can assay 8 to 10 samples per day by this method.

INTRODUCTION

DIOSGENIN is used in the synthesis of steroidal drugs. Several analytical procedures are available for the estimation of diosgenin in pure state or in *Dioscorea* tubers. Gravimetric procedure[1], gas liquid chromatography[2], colorimetry[3], densitometric thin-layer chromatography[4] and infra-red spectrometry[5] have been used. These methods involve more time, labour and material. The present paper describes a rapid method combining hydrolysis and extraction of *Dioscorea* tubers with spectrophotometric method for the determination of diosgenin in the extracts. The distinguishing features of this method are (*a*) small quantities (50 mg) of the dried plant material required, (*b*) hydrolysis and extraction achieved in 1½ to 2 hours, (*c*) avoiding additional solvents[3-6] and (*d*) use of smaller quantities of antimony ion for the colour reaction.

EXPERIMENTAL

Pure diosgenin was isolated from *Dioscorea deltoidea* and purified by preparative T.L.C. using $SbCl_5$ solution. (24% in 70% $HClO_4$) for detection.

Procedure

(*i*) *Preparation of Standard Curve.*—25 mg of pure diosgenin were dissolved in 250 ml hexane (Solution A) and 0·75 ml and 1·0 ml of this solution were taken in two tubes and the solvent removed by bubbling air. $HClO_4$ (5 ml) followed by 0·1 ml of $SbCl_5$ solution. (24% in 70% $HClO_4$)

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* To whom all inquiries should be addressed.

were added to each tube. The mixtures were kept for half an hour. Absorbance was noted for both the solutions from 400 to 600 nm against reagent blank. A maximum absorption was noticed at 486 nm.

Volumes corresponding to 10, 20, 30, 40, 50, 80 and 100 μg of diosgenin were taken from solution A into different tubes, solvent removed and the colour developed as mentioned above. The absorbance of each was noted at 486 nm against reagent blank.

(*ii*) *Hydrolysis and Extraction*.—Finely powdered tuber was dried at 105° C and passed through 72 mesh sieve. This material (50 mg) and 25 ml each of 3 N HCl and hexane were refluxed in an all-glass apparatus on a magnetic stirrer hot plate for 2 hours. The temperature of the liquid was maintained between 90–96° C. The mixture was allowed to cool and the aqueous phase was given two shakings with 25 ml of hexane each time and combined organic phase was washed with 1% $NaHCO_3$ solution and subsequently with distilled water and the volume made upto 100 ml with hexane. From suitable aliquots of this extract (usually 1 ml) solvent was removed and the colour developed as described above and the absorbance measured at 486 nm against the reagent blank. Gravimetric procedure[1] was also undertaken to find out the accuracy and efficiency of this method.

RESULTS AND DISCUSSION

Pure diosgenin when treated with $SbCl_5$ (24% in 70% $HClO_4$) showed λ_{max} at 486 nm. The red colour had a maximum absorbance after half an hour (23°–27°) and was stable for nearly two hours.

A straight line was obtained when the concentration of pure diosgenin (10, 20, 30, 40, 50 and 80 μg) was plotted against absorbance (Fig. 1). The colour reaction obeys Beer's Law at room temperature.

In the colorimetric procedure[3] for the estimation of pure diosgenin by a solution of $SbCl_3$ in a mixture of nitrobenzene and methanol, the solution is to be heated for at least 20 minutes at 60° C to develop the colour. Sofowora and Hardman[6] have developed a spectrophotometric method for the estimation of diosgenin in crude plant extracts. The procedure for obtaining crude plant extract has also been described[7]. In this method it takes more than 40 hours to obtain the crude plant extract which includes hydrolysis of the tuber, elimination of the acid, drying and extraction. Moreover, the stability of the red colour varies with time[6]. Reagents and tuber also required in larger quantities.

In the present method, only 50 mg of the dried material are adequate for the estimation. Both hydrolysis and extraction can be completed in about 2 hours. The quantitative aspect of the extraction has been confirmed by TLC of the aqueous layer. Hydrolysis conditions were thoroughly checked to make certain that the release of diosgenin was complete. In order to speed up the assay, advantage was taken of the immiscible character of hexane with aqueous phase. Hexane was found to be the most suitable of all the solvents tried. Only 0·1 ml $SbCl_5$ (24% sotution in 70% $HClO_4$) was found to give more stable colour as compared with 5 ml $SbCl_3$ (25% in $HClO_4$) used elsewhere[6].

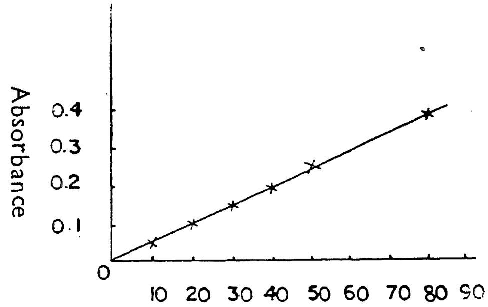

FIG. 1. Diosgenin concentration (μg).

The analytical results obained by gravimetry[1] and by the present method were nearly the same within the limits of experimental error (standard error = 0·15). The Students' *t* Test confirmed these observations. By the present method the analysis of 8 to 10 samples can be completed by one analyst in a day.

ACKNOWLEDGEMENTS

The authors are grateful to Prof. T. S. Sadasivan, Consultant to C.S.I.R., Plant-Based Laboratories, for his keen interest in this work. Thanks are also due to Dr. C. K. Atal, Director, Regional Research Laboratory, Jammu/Srinagar, for his valuable suggestions and to Dr. B. K. Bhat, for providing the facilities.

1. Morris, M. P., Roark, B. A. and Cancel, B., *J. Agric. Fd. Chem.*, 1958, **6**, 856.
2. Cooke, B. K., *Analyst*, 1970, **95**, 95.
3. Yamagishi, M. and Nakamura, I., *Yakugaku Zasshi.*, 1958, **78**, 1068.
4. Brain, K. R. and Hardman, R., *J. Chromat.*, 1968, **38**, 355.
5. Wall, M. E., Eddy, C. R., McClennan, M. L. and Klumpp, M. E., *Anal. Chem.*, 1952, **24**, 1337.
6. Sofowora, E. A. and Hardman, R., *Planta Medica*, 1974, **26**, 385.
7. Brain, K. R. and Hardman, R., *Phytochem.*, 1968, **7**, 1815.

LETTERS TO THE EDITOR

ON THE UNITARY ANALOGUE OF THE EULER TOTIENT FUNCTION

LET t be a fixed non-zero integer and $P_1, \ldots, P_r$ some arbitrary but fixed primes coprime to t. By Dirichlet's theorem there exist infinitely many primes p of the form $t + P_1 P_2 \ldots P_r A$ where A is a positive integer. Using this it is easy to see that if p is large,

$$\frac{\phi(p-t)}{\phi(p)} \leq 20 \prod_{i=1}^{r} \left(1 - \frac{1}{p_i}\right)$$

This proves by a proper choice of p_i that

$$\frac{\phi(p-t)}{\phi(p)} < \epsilon \text{ for all } p > p_e(\epsilon)$$

Corollary

$$\limsup \frac{\phi(n-t)}{\phi(n)} = \infty$$

$$\liminf \frac{\phi(n-t)}{\phi(n)} = 0$$

$$\limsup \frac{\phi^*(n-t)}{\phi^*(n)} = \infty$$

$$\liminf \frac{\phi^*(n-t)}{\phi^*(n)} = 0$$

where ϕ^* is the unitary analogue of the Euler's totient function (for definition of ϕ^* see the paper of Cohen[1]).

I am thankful to Dr. D. Suryanarayana, Andhra University, for suggesting the problem and to the referee for valuable suggestions in the presentation of this note.

Department of Mathematics,
B.J.B. College, Bhubaneswar-6,
Orissa, *October* 24, 1975.

B. S. K. R. SOMAYAJULU.

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1. Cohen, E., "Arithmetical functions associated with the Unitary divisors of an integer," *Mathematics Zeit.*, 1960, 74, 66.

ROTATIONAL (AND QUASI ROTATIONAL) BANDS IN Se, Xe, Ba AND Ce ISOTOPES

A CONSIDERABLE amount of information is now available regarding the rotational (and quasi rotational) bands in several isotopes of Se, Xe, Ba and Ce. We have examined the available data using a simple two parameter model[3], which includes at once rotations and vibration-rotation interactions to all orders. In this model the energy of level of spin J is

$$E_J = a[-1_J + \sqrt{1 + b^J (J + 1)}]$$

The values of the parameters a and b for the various isotopes are listed in Table I.

In the case of Se^{75}[1] there are two rotational bands one built on the ground state and another on an excited state. The parameters for the rotational band built on ground state of Se^{75} is different from these for other isotopes. The parameter a is very small and b is negative. Except for this a increases and b decreases with mass number.

In the case of xenon[2] the parameter a increases upto mass number 124 and afterwards gradually decreases whereas the parameter b decreases upto mass number 122 and then gradually increases.

In the case of cerium[2], the parameter a gradually decreases except between A = 132 and A = 134 isotopes whereas b gradually increases with mass number (*see* Table II).

As Z increases the parameter a increases and b decreases for the same mass number. (*see* Table II).

To draw complete benefit from this analysis it is necessary to have a good microscopic theory of rotations and rotation-vibration interaction. For rotational bands at least a good orientation is afforded by Elliott's SU$_3$ model according to which rotations may be attributed to pairing and quadru-

TABLE I
Selenium

Parameters/A	74	75	75	76
a	0·340787	0·0938947	0·423214	0·681626
b	1·19977	−0·3687920	0·423601	0·386349

TABLE II

Xehon, barium and cerium

Isotopes/A		120	122	124	126	128	130	132	134	136
Xe	*a*	0·513689	0·583774	0·584037	0·564995	0·485831	0·459017	0·389644	0·283195	
	b	0·274222	0·242671	0·264190	0·309502	0·443833	0·605515	1·070610	2·501950	
Ba	*a*			1·09277	1·0206	1·19041	0·682929	0·618969	0·493459	0·757879
	b			0·0771717	0·0940969	0·0886635	0·222026	0·349205	0·659208	0·564950
Ce	*a*					1·34748	1·066590	0·852115	0·860976	0·674259
	b					0·055225	0·0888326	0·151595	0·195958	0·384597

pole forces. Vibrations of nuclei can be regarded as superpositions of many particle-hole states. There seems to be no understanding yet regarding the role of the various features of nuclear interactions leading to the appearance of vibrations in nuclei. There seems to be no microscopic theory of vibration-rotation interaction. Hence we cannot say what parts of nuclear force are responsible for the observed behaviour of parameters *a* and *b*. It is even possible that features of nuclear force such as are not important from the point of view of the general nature of nuclear force are responsible for the above systematics of *a* and *b*.

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1. Zell, E. O., Friederichs, H. C., Heits, B., Von Brentano, P. and Protop, C., *Z. Physik*, 1975, 272 **A**, 27.
2. Wilhelmi, Z., *Fortschritte der Physik*, 1969, 17, 186.
3. Holmberg, P. and Lipas, P. O., *Nucl. Phys.*, 1968, **117 A**, 552.

pH METRIC STUDIES OF TARTRONIC ACID COMPLEX WITH METAL IONS

IN an earlier paper, we have reported the stability constants of tartronic acid with Cu (II), Ni (II) and Mn (II). In the present paper, are reported the complexing tendency of tartronic acid with metal ions such as Al (III), Fe (III), Mg (II), Ca (II) and Pb (II). The acid was used by 'Pavlinova[1], in a titrimetric method for the determination of aluminium. No systematic determination of the stability constants of the above metal ions with tartronic acid has been done, and hence the present investigation has been undertaken.

All the chemicals used in this work were of A.R. quality. All the solutions were standardised against EDTA[2] before use. For pH metric titrations the metal ion solutions were made in perchloric acid, the strength of the acid being determined titrimetrically[3]. A Philipps pH meter which has an accuracy of 0·05 pH unit was used for pH measurements, and it was standardised against potassium-hydrogen-phthalate and borax buffers before each experiment. All the experiments were performed at 27° C and ionic strength of 0·2 M.

TABLE I
pH titration data of the various complexes

Ionic strength	Metal								
0·200 M	Aluminium	pH	3·8	4·0	4·2	4·6	4·8	4·6	5·0
		$\bar{n}$	0·96	0·98	1·17	1·36	1·58	1·80	1·92
		pL	6·16	5·75	5·27	4·75	4·12	3·45	2·19
0·200 M	Magnesium	pH	3·2	3·8	4·0	4·4	4·8	5·4	
		$\bar{n}$	0·89	1·24	1·27	1·42	1·64	1·86	
		pL	7·39	6·02	5·61	4·71	3·70	2·89	
0·200 M	Calcium	pH	3·2	3·8	4·0	4·4	4·8	5·0	
		$\bar{n}$	0·99	1·24	1·27	1·49	1·80	1·97	
		pL	7·33	6·02	5·61	4·66	3·45	2·35	
0·200 M	Iron	pH	3·2	3·8	4·0	4·4	4·8		
		$\bar{n}$	0·96	1·35	1·39	1·63	1·72		
		pL	7·36	5·96	5·54	4·52	3·59		
0·200 M	Lead	pH	5·0	5·2	5·4	5·5	5·8	6·0	
		$\bar{n}$	0·53	0·75	0·92	1·20	1·50	1·84	
		pL	3·91	3·44	2·98	2·45	1·84	0·95	

Extermination of Stability Constants

Irving and Rossotti[4], described a procedure in which a mixture of the acidified solution of metal ion, and the ligand were titrated against alkali. Bjerrum's[5] pH titration technique has been applied in our present investigations. The method provides proton-ligand stability constants of the ligand, and those are employed for the calculation of the metal-ligand stability constants.

Proton-ligand stability constants have been calculated in our earlier paper (sent for publication) which gave the values:

$$\log K_1^H = 6.95 \text{ and } \log K_2^H = 2.10.$$

The solutions containing perchloric acid, perchloric acid + ligand and perchloric acid + ligand + metal solution, each diluted to a constant volume (50 ml) with conductivity water and maintained at constant ionic strength (0·2 M) by the addition of $NaClO_4$ were titrated against standard alkali. In all, three titrations were performed. From the titration curves, $\bar{n}$ and pL were obtained by Irving–Rossotti[4], method which are given in Table I.

From the values of $\bar{n}$ and pL the log K_1 and log K_2 have been calculated by use of mid point method[6] and interpolation at half $\bar{n}$ value method[6] respectively, which are given in Table II.

TABLE II
Stepwise stability constants of the various complexes

Metal ion	Log K_1	Log K_2	Log β
Al^{3+}	7·15	4·25	11·40
Mg^{2+}	10·05	4·55	14·60
Ca^{2+}	9·60	4·30	13·90
Fe^{3+}	9·15	5·05	14·20
Pb^{2+}	3·64	1·80	5·44

From Table II, it is seen that the stability of the metals increases in the order—Mg > Fe > Ca > Al > Pb.

Thanks are due to the U.G.C., for awarding a fellowship to one of us (S.M.).

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Indian School of Mines, NARENDRA S. RAWAT.
Dhanbad 826 004, *September* 12, 1975.

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1. Pavlinova, A. V., *J. Applied Chem.*, (U.S.S.R.), 1937, 10, 732; *C.A.*, 1937, 31, 6877.
2. Flaashka, H., *EDTA Titrations,* Pergamon, 1964.
3. Gran–G, *Analyst,* 1952, 77, 661.
4. Irving, H. M. and Rossotti, H. S., *J. Chem. Soc.*, 1954, p. 2904.
5. Bjerrum, T., *Metal Ammine Formation in Aqueous Solutions,* P. Haase, Copenhagen, 1941.
6. Irving, H. M. and Rossotti, H. S., *J. Chem. Soc.*, 1953, p. 3397.

USE OF SODIUM COBALTINITRITE AS A NEW SPRAY REAGENT FOR THE DETECTION OF SOME OPIUM ALKALOIDS ON TLC PLATES

THE detection of various opium alkaloids on TLC plates has been accomplished by a large number of spraying reagents and visualization techniques developed by various workers, which has been reviewed by Santavy[1].

Fiegel[2] has reported that in addition to phenolic compounds which have a free position ortho to a phenolic –OH group, morphine and many other compounds give a positive spot test with an aqueous solution of sodium cobaltinitrite. Bhatia *et al.*[3], have suggested a general spraying reagent for phenolic compounds on TLC plates which is a slight modification of the Fiegel's test. It was therefore thought worthwhile to investigate the efficacy of Bhatia *et al.*'s modified sodium cobaltinitrite reagent for the detection of five common opium alkaloids, *viz.*, morphine, codeine, papaverine, narcotine and thebaine, which is presented in this communication.

Thin layer plates of glass (20 × 20 cm) were prepared as described by Rama Rao and Murty[4] in which they have coated plates with silica gel—G—containing 4% sodium carbonate. The plates were activated at 110° C for one hour. The solvent system consisting of chloroform—ethanol (8 : 2) was used. The modified sodium cobaltinitrite reagent is prepared as described by Bhatia *et al.*[3], which is reproduced here. 4·4 g. $NaNO_2$ is dissolved in 10 ml water. To this solution a solution of CO $(NO_3)_2$ prepared by dissolving 2·6 g. CO $(NO_3)_2$ in 2 ml anhydrous acetic acid is added. To this combined solution 20 ml anhydrous acetic acid and 50 ml water is added. The plates were spotted with solutions of above alkaloids (1 mg/ml) prepared in chloroform (except morphine which was prepared in methanol). After development the plates were air dried and then sprayed with modified cobaltinitrite spray reagent. Subsequently after air drying the plates were heated in an oven at 105° C for 10 min. The colours observed under ordinary light and fluorescence observed under UV light along with the sensitivities are presented in Table I. The colours were stable for several hours. The initial blue background fades out gradually leaving the spots visible against a white background.

The reagent was tried on an extract of opium and was found to give good visualization. The reagent appears to be sensitive for visualizing the alkaloids of opium in crude extracts as well as in other samples of forensic interest.

TABLE I

Visualization of some common opium alkaloids on TLC plates with modified sodium cobaltniitrite spray reagent.

Alkaloid	Colour observed in daylight	Fluorescence under UV light	Minimum detectable limit (μg)
1. Morphin	Yellowish brown	Green	5
2. Codeine	do.	..	20
3. Thebainee	do.	..	5
4. Papaverine	do.	..	5
5. Narcotine	do.	Bluish green	5

The authors wish to thank Dr. S. N. Garg, for his keen interest and encouragement.

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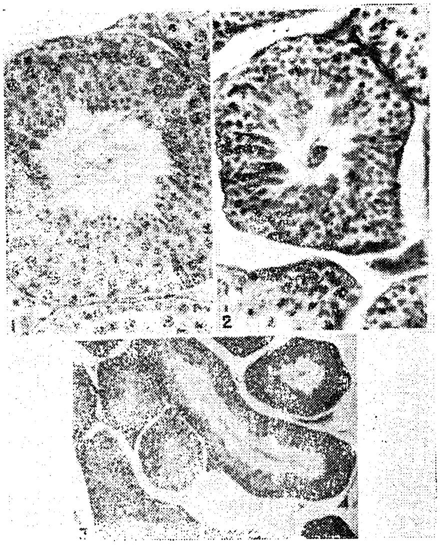
1. Santavy, F., in Stahl, E., (Ed.), *Thin Layer Chromatography*, A Laboratory Hand Book, Springer, New York, 1969, p. 436.
2. Fiegel, F., *Spot Tests in Organic Analysis*, Elsevier, New York, 1966, p. 183.
3. Bhatia, I. S., Bajaj, K. L., Verma, A. K. and Singh Joginder, *J. Chromatogr.*, 1971, 62, 471.
4. Rama Rao, N. V. and Murty, H. R. K., *J. Ind. Acad. Forens. Sci.*, 1974, 13, 18.

ORIENTATION OF THE SPERMATOZOA IN THE SEMINIFEROUS TUBULES OF SOME BATS

THE mechanism by which spermatozoa are trasported from their place of origin in the seminiferous tubules to the epididymis is not clearly understood and has led to considerable speculation by workers. Most of the workers however agree that the spermatozoa are passive in this process, being either carried by a fluid current through the seminiferous tubules or passed towards the distal regions of the tubules by undulating movements of the walls of the seminiferous tubules. A critical review of the earlier work on this subject is given by Bishop[1].

During the course of a study of the male reproductive structurec of several species of Indian bats the authors noted that bundles of sperm tails are oriented in a spiral manner in transverse sections of the seminiferous tubules during the breeding season in all the species of bats studied (Figs. 1 and 2). The sperm heads lie radially towards the periphery of the lumina of the seminiferous tubules. There seems to be a strong current within the seminiferous tubules, and since the sperm tails are lighter than the heads, the tails are drawn first into the current. Hence, in sections in which the tubules are cut longitudinally the tails of the sperms are all oriented in one direction and lie in the centre of the tubule (Fig. 3). The orientation of the tails in a spiral manner indicates that the current has a rotary movement inside the tubules.



FIGS. 1–3. Figs. 1 and 2. Transverse sections of the seminiferous tubules of *Rousettus leschenaulti* (Megachiroptera-Pteropidae) and *Pipistrellus ceylonicus chrysothrix* (Microchiroptera-Vespertilionidae) respectively. Note the spiral orientation of the sperm tails in the centre of the seminiferous tubules. The heads of the sperms are located towards the periphery of the lumen, × 280. Fig. 3. Part of the testis of *Rousettus leschenaulti* showing one tubule cut longitudinally and several cut transversely. The sperm tails are all oriented in one direction in the centre of the longitudinally cut tubule, × 60.

Two questions arise in this context : (*i*) where does the fluid in the seminiferous tubules come from, and, (*ii*) what propels this fluid through the tubules. It is suggested that the large quantity of cytoplasm which is sloughed off from the spermatids during spermateleosis, may form the major bulk of this fluid, and the cytoplasm of the degenerating sertoli cells may also contribute to this mass. The peristaltic movements caused by the alternate contraction and expansion of the wall of the seminiferous tubules (Roosen–Runge[3],) may be responsible for the propulsion of the fluid through the seminiferous tubules.

The mechanism responsible for bringing about a rotary movement of the fliud in the seminiferous tubules is not known. It is here pertinent to mention that the spermatozoa do not appear to have a specific orientation in stained sections of the epididymis (Gopalakrishna *et al.*[2]). Apparently, the rotary nature of the current is confined to the seminiferous tubules only.

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Institute of Science, K. B. KARIM.
Nagpur, *September* 21, 1975.

1. Bishop, D. W., In : *Sex and Internal Secretions*, Ed., W. C. Young, The Williams and Wilkins Co., Baltimore. 1961.
2. Gopalakrishna, A., Thakur, R. S. and Madhavan, A., *Curr. Sci.*, 1974, 43, 786.
3. Roosen–Runge, E. C., *Anat. Rec.*, 1951, 109, 413.

HOST-PATHOGEN EQUILIBRIUM BETWEEN RICE (*ORYZA SATIVA* L.) AND ITS PATHOGEN *PYRICULARIA ORYZAE* CAV.

THE first report of the occurrence of physiologic races in *Pyricularia oryzae* Cav. in India is by Padmanabhan[1]. Padmanabhan *et al.*[2], reported the occurrence of thirty-one races of the pathogen after studying the pathogenicity-patterns of one hundred and thirty-two isolates of *P. oryzae* obtained through isolation from blast specimens collected from different States of India. Occurrence of one more race of the pathogen was reported from Central Rice Research Institute[3]. Veeraraghavan and Premalatha Dath[4], could identify during 1972, the incidence of two races of the pathogen, *viz.*, IC 1 and IC 17 only[5]. The isolates included in that study were from disease specimens collected during 1971 and 1972. Isolates of *P. oryzae* which were collected during 1972 and 1973, revealed the presence of the race IC 17 in different parts of this country.

The present contribution reports the occurrence of physiologic races of *P. oryzae* which were prevalent during 1973 and 1974.

One hundred and ten isolates of the pathogen were obtained from blast specimens collected from Kerala, Tamil Nadu, Karnataka, Andhra Pradesh, Orissa, West Bengal, Tripura, Meghalaya, Assam, Manipur, Bihar, Madhya Pradesh, Uttar Pradesh, Himachal Pradesh, Punjab, Jammu and Kashmir, Rajasthan, Gujarat and Maharashtra. The international set of eight differentials[6], *viz.*, Raminad str.[3], Zenith, N.P. 125, Usen, Dular, Kanto-51, CI 8970 (S), and Caloro were utilised for the

TABLE I

Pathogenicity-pattern of the physiologic race IC 17 Pyricularia oryzae *Cav.*

Raminad str. 3	Zenith	N.P. 125	Usen	Dular	Kanto-51	CI 8970(S)	Caloro
R	R	S	R	S	S	S	S

R = Resistant, S = Susceptible.

identification of physiologic races of the pathogen. The highly susceptible variety, Co. 13 was utilised as a check for detecting the success of inoculation and the identity of the physiologic race of the pathogen which may not produce any visible symptoms on the plants of the eight differentials[2]. The method of identification of the physiologic races of *P. oryzae* was the one described by Padmanabhan *et al.*[7]. The classification of Ling and Ou was adopted[5].

The data revealed that the one hundred and ten isolates of the pathogen belonged to the race IC 17[5]. The pathogenicity-pattern of this race is presented in Table I.

The physiologic race IC 17[5] was reported to be the most prevalent one in India since 1962[2,4]. The predominant prevalence, survival and succession of the race IC 17 over the other races indicates that this is a very stable race.

Miura and Ito[8], have reported the 'prosperity and decline of the race C in Japan'. Goto[9], stated "a certain delay is expected in the increase of adapted (or severe) races as compared with the increased cultivation of the varieties with new resistance; for instance, a delay lasted about ten years in the case of the variety, Futaba". The time lag between the introduction of a resistant variety and the break-down of resistance in the plants of the variety—obviously by the appearance of a new physiologic race of the pathogen—denotes a phase of host-pathogen equilibrium in the area. This phenomenon is evidenced by the monoracial succession brought about by the elimination of less stable biotypes of the pathogen and non-appearance of new races of the pathogen in the area. During the period under report no instance of break-down of resistance in a rice variety on a field scale has come to the notice of the authors, which has been known for its resistance in that locality.

The authors are thankful to Dr. S. Y. Padmanabhan, Director, for constant encouragement and keen interest. Help rendered by Sri. N. K. Chayau Patnaik, and Sri Rama Ranjan Das, is also gratefully acknowledged. The supply of the blast specimens by various organizations is gratefully acknowledged.

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---

1. Padmanabhan, S. Y., *Curr. Sci.*, 1965, **34**, 307.
2. —, Chakrabarti, N. K., Mathur, S. C. and Veeraraghavan, J., *Phytopathology*, 1970, **60** (11), 1574.
3. *Annual Report*, Central Rice Research Institute, Cuttack, India, 1969, p. 109.
4. Veeraraghavan, J. and Premalatha Dath, A., *Curr. Sci.*, 1975, **44** (1), 19.
5. Ling, K. C. and Ou, S. H., *Phytopathology.*, 1969, **59**, 339.
6. Atkins, J. G. *et al.*, *Ibid.*, 1967, **57**, 297.
7. Padmanabhan, S. Y., Chakrabarti, N. K., Mathur, S. C., Veeraraghavan, *J. Proc. Ind. Acad. Sci.*, 1967 **76 B**, 63.
8. Miura, H. and Ito, H., *Bull. Yamagata Prefect. Agric. Exp. Stn.*, 1974, **8**, 92.
9. Goto, K., In: *The Rice Blast Disease*, John Hopkins Press, Baltimore, Maryland, U.S.A.. 1965, p. 237.

SALT TOLERANCE OF COTTON AND POTENTIAL USE OF SALINE WATER FOR IRRIGATION

A SATISFACTORY stand of cotton crop on saline soils and/or under saline water irrigation is a serious problem of common occurrence. Crop failures are due to the accumulation of salts which results in the reduction of seed germination and plantgrowth. Cotton, being recognised as the most salt tolerant of all the field crops, has not received the salinity study. The present work deals with the relative salt tolerance of some important cotton varieties and hybrid Varalaxmi at germinatios stage with different levels of salinity under field conditions.

The experiment was conducted in bottomless coal tar drums embedded in medium black clay soil (vertisol). Bhagya, Hampi, Laxmi and hybrid Varalaxmi were used in the present investigation. Saline water used for irrigation (conductivity values of 4, 8 and 12 mmhos/cm) was prepared by dissolving sodium chloride, sodium sulphate, sodium bicarbonate and calcium chloride on weight proportions (w/w) so as to obtain Na : Ca in the ratio of 4 : 1 and C_1 : SO_4 : HCO_3 in the ratio of 2 : 1 : 1[1]. Two replications were maintained for each treatment. Saline water was added and the soil was incubated for 15 days prior to sowing. Ten seeds were sown in each drum and the germination counts were recorded till 30th day. Plant samples were then drawn and dried at 70° C and used for chemical analysis. The leaf samples were analysed for sodium and potassium[3].

The data on germination recorded on 10, 20 and 30th day after sowing reveal that hybrid Varalaxmi had better germinability. At the salinity level of 12 mmohs/cm, per cent germination recorded on 30th day was 40 and 25 in the case of Varalaxmi and Bhagya and 15 in the case of both Hampi and Laxmi. These differences may have greater impact, if cotton has to be grown on a salt affected land and/or under saline water irrigation. Thus, selection of a climatically adopted salt tolerant variety and/or hybrid rather than one of lower salt tolerance may mean the difference between a good to a fair stand.

The effect of presowing salinization on cationic composition of cotton leaves are presented in Table I.

Presowing salinization affected the uptake of Na and K. Leaf samples of all the varieties revealed a sharp increase in Na content with increasing salinity of the water and support the findings of other workers[1,4,5,8]. Sodium uptake by Varalaxmi and Bhagya was much lower than in Hampi and Laxmi in relation to its abundance and availability in the applied saline waters. This strongly suggests that Varalaxmi and Bhagya exclude sodium absorption by some selective mechanism. This exclusion of sodium may partly be responsible for the relatively high tolerance shown by Varalaxmi and Bhagya and this is common to many salt tolerant plants[4,6-8].

In the present study K : Na ratios as high as 1·6 in Varalaxmi and Bhagya as compared to a lower K : Na ratio of 0·63 in Hampi and 0·45 in Laxmi at the salinity level of 12 mmhos/cm provide support to the opinion that K : Na ratio has a role[5,10] in determining salt tolerance in plants (Table I).

Since this crop is grown primarily for its seed cotton, the effect of presowing irrigation with saline water on seed cotton yield is of importance. The per cent drop in seed cotton yield at the salinity level of 12 mmhos/cm compared to the yield obtained under best available water (conductivity of 1 mmhos/cm) used for irrigation was 9. 45, 59 and 75 in the case of Varalaxmi, Bhagya, Laxmi and Hampi respectively.

The ability to exclude sodium in relation to its relative data availability in the applied saline waters, the data on germination and seed cotton yield indicate the salt tolerance potential of Varalaxmi and Bhagya.

TABLE I

Ionic composition of cotton leaves as affected by presowing salinization

Ionic constituents, meq/100 g of dry matter	Salinity level, mmhos/cm	Varieties/Hybrid			
		Varalaxmi	Bhagya	Hampi	Laxmi
Na	0	4·17	3·83	7·48	6·87
	4	6·96	19·14	26·53	23·49
	8	26·97	29·58	28·70	41·33
	12	28·72	16·53	59·16	65·25
K	0	48·58	44·74	42·51	34·80
	4	46·02	35·74	28·38	35·79
	8	54·97	42·19	38·99	42·19
	12	48·24	27·29	37·07	29·12
K:Na ratio	0	11·66	11·68	5·68	5·06
	4	6·61	1·87	1·07	1·52
	8	2·04	1·43	1·36	1·02
	12	1·68	1·65	0·62	0·44

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---

1. Anonymous, *Proceedings of Combined Fifth Annual Work Shop on Water Management and Salinity*, 1973.
2. Chang, C. W. and Dregne, H. E., *Soil Sci. Amer. Proc.*, 1955, **19**, 29.
3. Chapman, H. D. and Pratt, P. F., *Methods of Analysis for Soils, Plants and Waters*, Univ. of California, Riverside, 1961.
4. Hayward, H. E. and Wadleigh, C. H., *Advances in Agronomy*, 1949, **1**, 1.
5. *Irrigation, Drainage and Salinity*—An International Source Book, 1973, FAO/UNESCO, 510 pp.
6. Lagerwerff, J. V., *Soil Sci.*, 1958, **86**, 63.
7. Larson, W. D. and Pierre, W. H., *Ibid.*, 1953, **51**, 51.
8. Longenecker, D. E., Thaxton, E. L. and Lyerly, P. J., *Texas Agri. Expt. Sta. Misc. Publ.*, 1964, p. 728.
9. Pearson, G. A. and Bernstein, L., *Soil Sci.*, 1958, **86**, 254.
10. Rodney, R. B., Boswell, S. B. and Whiting, F. L., *Proc. Amer. Soc. Hort. Sci.*, 1956, p. 234.

DEXTRANASE PRODUCTION BY CERTAIN PLANT PATHOGENIC FUNGI

Dextranase, which hydrolyses α-1, 6 glucosidic linkages in dextran, is of common occurrence in moulds and bacteria[1,2]. Though amylases are secreted by fungi they are reported not to hydrolyse any bonds in dextran even though the type of glucosidic linkage in dextran and starch is similar. Three soil-borne plant pathogenic fungi, *viz.*, *Rhizoctonia bataticola*, root-rot fungus of peanut, *Sclerotium oryzae*, stem-rot fungus of rice and *Fusarium oxysporum* f. *melonis*, the muskmelon wilt fungus were tested for the production of extra cellular dextranase and the results are reported here.

Czapek's medium containing dextran at 3% level as carbon source was dispensed in 50 ml quantities into 250 ml Erlenmeyer flasks, sterilized at 15 lb psi and inoculated with 8 mm disc of the fungi. Replicates were maintained for each organism. After 15 days of inoculation the biomass was filtered through previously oven dried and weighed filter-paper, dried at 105° C for 24 hr and reweighed. The dry weights of the biomass were recorded separately. The culture filtrates were retained for enzymes assay. The assay method described by Tsuchiya *et al.*[2], was followed with a modification for determining the dextranase activity. The reaction mixture contained 4 ml of 1% dextran suspended in sodium acetate-acetic acid buffer (pH 5·1), 2 ml of the buffer and 2 ml of the enzyme source. The release of the reducing sugar was estimated at 0 and 2 hr[3]. The activity of the enzyme on starch was also investigated.

The biomass production and enzyme activity of the three fungi are reported in Table I.

Table I

Effect of dextran on growth and dextranse production of the three fungi

Fungus	Biomass (mg)	Enzyme activity* (mg/ml)	
		On Dextran	On Starch
Rhizoctonia bataticola	250·0	0·223	0·295
Sclerotium oryzae	104·5	0·017	..
Fusarium oxysporum f. *melonis*	200·5	0·038	0·030

* Expressed as glucose equivalents.

Although Bourdu and Quilet[4] have reported the occurrence of dextran in the roots of *Anchusa sempervirens*, the substance, however, has not been located on most plant species[5]. Dextranase production by plant pathogens has not so far been reported. In the present study, of the three pathogens *R. bataticola* exhibited a marked dextranase activity and recorded a higher biomass followed by *F. oxysporum* f. *melonis* and *S. oryzae*. Interestingly, the culture filtrate from *R. bataticola* hydrolyzed starch effectively while that of *F. oxysporum* f. *melonis* showed only traces of activity. However, the enzyme from *S. oryzae* failed to act upon starch. Repression or inhibition of pathogen's enzyme(s) by fungitoxicants of the host, such as phenols was reported earlier[6,7,8]. The ability of *R. batatiola* and *F. oxysporum.* f. *melonis* to elaborate dextranase that can also act upon starch might therefore be of importance in pathogenesis as the dextranase might be active in case of amylase inhibition or repression by fungitoxicants of the host. Extracellular dextranase production by certain saprophytic fungi is well known[2]. All the three plant pathogens in the present study are soil-borne. Dextran is present in soil through the synthesis by soil bacteria[9]. Production of dextranase by these soil-borne plant pathogens might therefore aid them in utilising dextran as a substrate in their saprophytic survival.

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---

1. Serry, T. W. and Hehre, E. J., *J. Bact.*, 1956, 17, 373.
2. Tsuchiya, H. M., Jeanes, A., Bricker, E. M. and Wilham, G. A., *J. Bact.*, 1952, 64, 513.
3. Nelson, A., *J. Biol. Chem.*, 1944, 153, 375.
4. Bourdu, R. and Quilet, M., *Compt. Rend.*, 1953, 237, 1751.
5. Swanson, M. A., *J. Biol. Chem.*, 1948, 172, 805.
6. Koti Reddy, M., *Annamalai Univ. Agri. Res. Annu.*, 1970, 2, 53.
7. — and Mahadevan, A., *Indian Phytopath.*, 1967, 20, 265.
8. Mahadevan, A. and Koti Reddy, M., *Neth. J. Pl. Path.*, 1968, 74, 87.
9. Quastel, J. H., "Microbial activities of soil as they affect plant nutrition," In: *Plant Physiology A Treatise*. Ed. by F. C. Steward, 1963, 3, 671.

FATTY ACID OXIDATION BY THE LEG MUSCLES OF *CYBISTER* BEETLE DURING VIGOROUS EXERCISE

IT is well recognized now that some insects are capable of utilizing fatty acids during their vigorous muscular activity as in flight[1-2]. Earlier studies have revealed that the flight muscles of some insects are capable of oxidizing only short chain fatty acids—butyrate and octanoate but not higher chain fatty acids—palmitate and stearate[3-4]. George and Bhakthan[5], observed that the flight muscle homogenate of the honey bee *Apis dorsetta* was capable of oxidizing butyrate.

That added carnitine stimulates fatty acid oxidation is now very well understood[6-7]. The aquatic dytiscid beetle *Cybister confusus* has an efficient leg muscle system. It is possible therefore that the leg muscles have an efficient biochemical machinary to meet the exigencies of energy demands, for all its spasmodic actions. The present investigation is aimed at ascertaining the ability of the leg muscles of the *Cybister* beetle to oxidize various fatty acids during its vigorous exercise.

The adult *Cybister* beetles, were vigorously exercised following the method described elsewhere[8]. Muscle homogenate and mitochondrial preparations were obtained as outlined in an earlier study[7]. Sodium salts of butyrate, octanoate, palmitate and stearate were used as substrates for fatty acid oxidation. The oxygen uptake by the different preparations was expressed as μl per mg. protein per 30 min. The procedure followed was essentially similar to that described elsewhere[7]. The effect of added carnitine on palmitate oxidation was tested in the muscle homogenate as well as in the mitochondrial preparation with 0·15 M DL-carnitine. The protein contents of the homogenate and mitochondrial fraction were determined according to the method of Lowry *et al.*[9].

The respiratory measurements obtained by the unexercised and exercised leg muscles homogenate preparations (Table I) after the addition of the various fatty acids revealed that they are capable of oxidizing only short chain fatty acids butyrate and

TABLE I

Oxidation of butyrate, octanoate, palmitate and stearate by the exercised and unexercised leg muscle homogenates of the Cybister *beetle*

Substrates	O_2 uptake (μl/mg. protein/30 min.)	
	Unexercised	Exercised
Endogenous	38	46
Butyrate	49	55
Octanoate	45	55
Palmitate	37	37
Stearate	36	37
Succinate*	70	103

The results presented in Tables I and II are averages of three readings in each case.
* Used for comparison.

TABLE II

Effect of added carnitine on the palmitate oxidation by the leg muscle homogenate and mitochondrial fraction of the Cybister *beetle*

Substrates	O_2 uptake (μl/mg. protein/30 min.)	
	Homogenate	Mitochondrial pellet
Endogenous	39	32
Palmitate added	37	33
Carnitine added	45	39
Carnitine + Palmitate added	47	50

octanoate but not the higher chain fatty acids—palmitate and stearate even when the leg muscles were metabolically active (as indicated by succinate oxidation in the exercised muscles). That the butyrate acts as primer in the oxidation of higher chain fatty acids in the locust thoracic muscles[10], and also as energy source during flight in the flight muscles of the honey bee *Apis dorsetta*[5] have been very well demonstrated. It has also been shown that the flight muscles of the dragonfly *Pantala flavescens*[7], which are involved in sustained action utilize short chain fatty acids to begin with, and subsequently switch on to its reserves of palmitic acids. It may be inferred from these observations that the leg muscles of this beetle which take part in short and abrupt actions derive their metabolic energy through limited oxidation of low chain fatty acids—butyrate and octanoate.

It is well understood now that carnitine stimulates the rate of oxidation of fatty acids both in the homogenate as well as in the mitochondrial fraction[11]. In the present investigation also palmitate oxidation in the homogenate as well as in the mitochondrial preparation was indicated after the addition of carnitine. This may suggest that in the leg muscles of this insect carnitine acts as an extra mitochondrial factor accelerating palmitate oxidation. It may be that the leg muscles of the *Cybister* beetle have insufficient carnitine reserves to support palmitate oxidation.

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---

1. Weis-Fogh, T., *Phil. Trans. R. Soc. London*, 1952, **237**, 1.
2. Beenakkers, A. M. Th., *J. Insect. Physiol.*, 1965, **11**, 879.
3. Rees, K. R., *Biochem. Z.*, 1954, **58**, 196.
4. McShan, W. A., Kramer, S. and Schlegel, V., *Biol. Bull. Wood's Hole*, 1954, **106**, 341.
5. George. J. C. and Bhakthan, N. M. G., *J. Insect. Physiol.*, 1963, **9**, 311.
6. Bode, C. and Klingenberg, M., *Biochem. Z.*, 1965, **341**, 271.
7. Kallapur, V. L. and George, C. J., *J. Insect Physiol.*, 1973, **19**, 1035.
8. —, *Curr. Sci.*, 1973, **42** (7), 254.
9. Lowry, O. H., Rosebrough, W. J., Farr, A. I. and Randall, R. J., *J. Biol. Chem.*, 1951, **193**, 265.
10. Meyer, H., Preiss, B. and Bayer, Sh., *Biochem. J.*, 1960, **76**, 27.
11. Fritz, I. B. and Marquis, N. R., *Proc. Natn. Acad. Sci. U.S.A.*, 1965, **54**, 1226.

CHANGES IN THE CORPUS ALLATUM VOLUME OF FEMALE *SPODOPTERA LITURA* (FABR.) RELATED WITH AGE AND REPRODUCTION

The importance of the corpus allatum in growth and reproduction of insects was first demonstrated in *Rhodnius prolixus*[1] (Hemiptera : Heteroptera). Since then it has been confirmed in a number of species belonging to Orthoptera, Hemiptera, Coleoptera, Lepidoptera and Diptera. Several workers reported larger corpora allata in females than in males of different species and emphasized its role in oocyte growth and yolk deposition during the maturation of eggs. Further, in *Calliphora erythrocephala*[2,3], *Sarcophaga bullata*[4], *Iphita limbata*[5], *Leucophaea maderae*[6], *Locusta migratorea*[7] and *Schistocerca* sp.[8], the increase in the volume of corpora allata of the females was related with oocyte growth and the maturation of the eggs in different reproductive cycles. But in moths, except *Bombyx mori*[9], males have larger corpora allata than females and gland was not necessary for the ovarian maturation. On the contrary, in butterflies *Pieris brassicae*[10], *P. napi*[11] and *Danaus plexippus*[12], studied so far, the corpora allata are larger in females than in males and these are necessary for the oocyte growth and egg maturation.

The present observation on the female *Spodoptera litura*, not only compares the volume of corpora allata with that of other moths but also indicates the changes in their volume related with age and reproduction. Further, the present data on *S. litura* particularly add information on the role of corpora allata in the short lived moth which requires food in imaginal life unlike *B. mori* and others which emerge only to lay eggs.

From a stock culture maintained at 27° C ± 1° C and 70–80% relative humidity, newly emerged *Spodoptera litura* were isolated in single pairs and maintained age-wise at the above-mentioned temperature and humidity. Their corpus allatum volume was determined by the method of Highnam (1958)[13]. The mean values of the corpus allatum volume of the females of different age groups was compared and significant difference was known by using the formula of Rao (1952)[14].

The data in Table I show the changes in the corpus allatum volume of the females of different ages. The volume of the gland was also determined in the newly emerged males and its mean value ($5\cdot 47 \pm 0\cdot 36 \times 10^6\mu^3$) was lower than that of the females of the corresponding age ($5\cdot 51 \pm 1\cdot 26 \times 10^6\mu^3$). However, in the females of the subsequent age, the gland volume significantly decreased and in the four day old females this volume was almost half of that of the newly emerged females. When the females were starved from emergence, the corpus allatum volume of the females of different age was significantly lower than that of the fed females of the corresponding age.

TABLE I

Showing changes in the corpus allatum volume of female Spodoptera litura *following emergence*

Age of female following emergence	Volume of corpora allata in $10^6 \mu^3$ (Mean value)	
	Fed	Unfed
Newly emerged	$5 \cdot 51 \pm 1 \cdot 26$	..
One day	$3 \cdot 90 \pm 0 \cdot 53$	$3 \cdot 46 \pm 0 \cdot 46$
Two day	$3 \cdot 30 \pm 0 \cdot 51$	$2 \cdot 96 \pm 0 \cdot 54$
Three day	$2 \cdot 75 \pm 0 \cdot 31$	$2 \cdot 63 \pm 0 \cdot 59$
Four day	$2 \cdot 63 \pm 0 \cdot 43$	$2 \cdot 42 \pm 0 \cdot 14$

The dimorphism in the corpus allatum volume of the newly emerged adults suggests that the gland of the newly emerged females have more cells and higher quantity of secretory material both in extra- and intracellular spaces of the gland. The subsequent decrease in the volume of the corpus allatum of fed females following emergence indicates gradual retardation in the secretory activity of the gland. Further, starvation following emergance markedly reduces the activity of the corpus allatum.

Anatomical observations of the ovary of *S. litura* showed that in the newly emerged females the anterior one-third of the ovarioles had immature eggs with still large nurse cells. Further, the commencement of oviposition of eggs in *S. litura* occurs after 60 to 70 hours following emergence. Therefore, the large size of the corpora allata of the newly emerged females of *S. litura* is most likely related with the synthesis and release of the secretory material for the completion of the maturation of the growing oocytes.

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---

1. Wigglesworth, V. B., *Quart. J. micr. Soi.*, 1936, **79**, 91.
2. Thomsen, E., *Vidensk. Medd. dansk. naturh. Foren. Kbh.*, 1942, **106**, 319.
3. Lea, A. O. and Thomsen, E., *J. Insect Physiol.*, 1969 **15** (3), 477.
4. Wilkens, J. L., *Ibid.*, 1968, **14**, 927.
5. Nayar, K. K., *Quart. J. micr. Sci.*, 1956, **97**, 83.
6. Scharrer, B.. *Biol. Bull. Wood's Hole*, 1952, **102**, 261.
7. Highnam, K. C. and Haskell, P. T., *J. Intect Physiol.*, 1964, **10**, 849.
8. Strong, L., *Ibid.*, 1965, **11**, 135.
9. Ito, H., *Bull. imp. seru. Coll. Tokyo*, 1918, **1**, 63.
10. Karlinsky, A., *C. R. Acad. Sci. Paris*, 1963, **256**, 4101.
11. Palm, N. B., *K. Svenska Vetensk Acad. Handl.*, 1949, **4**, ser. I (6), 24.
12. Barker, J. F. and Herman, W. S., *J. expt. Zool.*, 1973, **183** (1), 1.
13. Highnam, K. C., *Quart. J. micr. Sci.*, 1958, **99**, 171.
14. Rao, C. R., *Advanced Statistical Methods in Biometric Research*, John Wiley and Sons, New York, 1952.

ON THE OCCURRENCE OF TWO NEW PARASITIC NEMATODES, *ECHINOCEPHALUS SPINOSISSIMUS* (LINSTOW, 1905) AND *E. UNCINATUS* IN THE EAGLE RAY, *AETOBATIS NARINARI* (EUPHRASAN) FROM BAY OF BENGAL

IN the course of a survey of the parasites of commercially important elasmobranchs, a careful examination of the guts, sampled on two different occasions, yielded specimens of two species of nematodes. These nematodes were recovered from the intestines of *Aetobatis narinari* (Euphrasan), a common eagle ray of the region. The specimens were assigned to the genus *Echinocephalus* because of the presence of the generic characters such as the head bulb with transverse rows of hooks and the neck glands[1]. Subsequently they were identified as *Echinocephalus spinosissimus* (Linstow, 1905) and *E. uncinatus* (Molin, 1858). The specimens have been deposited in the Marine Biological Station Reference Museum collection.

As these parasites were collected from a hitherto unreported host and since *E. spinosissimus* has not so far been reported from Indian waters, the present communication adds further information and a brief description.

Echinocephalus spinosissimus (Linstow, 1905)

Material: Three males, 14 mm; 15 mm and 16·5 mm in length collected on October 1, 1971, *Host and Locality* : *Aetobatis narinari* (Euphrasan), entire fish weighing 30 kg. Bay of Bengal, Porto Novo (11° 29′ N 79° 49′ E).

Location : Inner wall of the intestine.

Three specimens ranging from 0·83 to 1·03 mm in width were collected from the intestine of the host. Body linear, striated with six terminal lips. Head bulbs well developed with 27 rows of hooks. Four thin neck glands, and eight pairs of minute papillae with a well-developed proximal pair present on sides of body.

Echinocephalus uncinatus (Molin, 1858)

Material : One male, 13 mm in length collected on December 10, 1971.

Host and Locality : *Aetobatis narinari* (Euphrasan). 24 kg in weight ; Bay of Bengal, Porto Novo.

The nematode was found attached to the inner wall of the stomach, coiled up in an ovoid cyst. Body smooth with no trace of annulations and measured 0·5 mm in width ; head bulb with 6 transverse rows of hooks. Neck glands straight and devoid of swellings.

E. spinosissimus (Linstow, 1905) has so far been reported only from Ceylon waters[2,3]. Molin[2] first described the species from *Trygon* spp. under the name *E. uncinatus* and later Linstow[3] reported the occurrence of this species from *Myliobatis aquila* and *Trygon* spp., but under the name *Cheirocenthus spinosissimus.* This species has not so far been reported from Indian waters.

E. uncinatus has been reported to occur in *Trygon* and *Myliobatis*[2,6,5] in gastropods[6] and in Scianeid fishes[7], in Indian waters. The present records its occurrence from a new host, *viz.*, *Aetobatis narinari* which probably forages on gastropods. In this context it may be relevant to refer to Anantaraman's[6] report on the occurrence of *E. uncinatus* in marine gastropods. Possibly the marine gastropods serve as the intermediate hosts.

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Tamil Nadu, *March* 1, 1976.

---

1. Yamuguti, S., "Systema helmitum. III," *The Nematodes of Vertebrates* (Interscience Publication, Inc., N.Y.), 1961, Parts 1 and 2.
2. Molin. R., *Wein. Math–Naturw. C.I.*, 1858, 28, 365.
3. Linstow, W., *Report to the Government of Ceylon on the Pearl Oyster Fisheries of the Gulf of Mannar* (Herdman, London), 1904, p. 2.
4. Baylis, H. A. and Lane, C., *Proc. Zool. Soc. London*, 1920, p. 245.
5. Shipely, A. E. and Hornell, J., *Report to the Government of Ceylon on the Pearl Oyster Fisheries of the Gulf of Mannar* (Herdman, London), 1905, 3.
6. Sita Anantaraman, *Studies of Helminths of Madras, Ph.D. Thesis,* University of Madras, India, 1961.
7. Muthiah, P., *Parasites of Sciaenid Fishes from Bay of Bengal, off Porto Novo,* M.Sc. Thesis, CAS in Marine. Biolovy, Annamalai University, India, 1972.

GIANT CELLS FORMATION IN THE TESTES OF HOUSE SPARROW *PASSER DOMESTICUS* (LINN.) BY COBALT-60 EXTERNAL IRRADIATION

It has been observed that irradiation causes the formation of giant cells in the testes of mammal[1-3]. As there is paucity of such information in birds, the present investigation is to study the effect of Cobalt-60 irradiation on the testes of the House Sparrow *Passer domesticus.* The whole body of these birds was exposed to external irradiation of Cobalt-60 at varying doses of 475 r, 712 r and 950 r at the rate of 100 r/h from a distance of 1 foot and autopsied on days 1, 2, 3, 7 and 15. The testes were fixed in Bouin's fluid, sectioned at 5 μ thickness and stained with Ehrlich's haematoxyline and PAS for histological observations.

The giant cells could not be observed after 475 r dose, but with the increase in the dose, the cells began to cluster and at 950 r many bi- and multinucleated giant cells were located. The clustering phenomenon appeared to start when the testes were irradiated by 712 r and autopsied on 3rd day, when many cells began to coalesce and formed binucleated or multinucleated giant cells having a common cytoplasmic pool (Fig. 1).

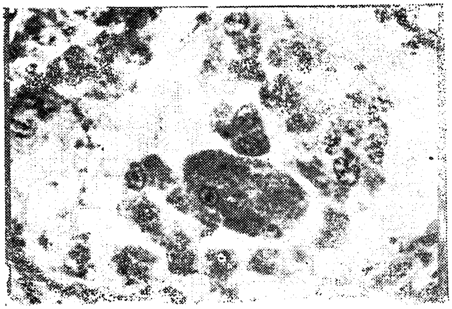

Fig. 1. Seminiferous tubule of the testes of male house sparrow *Passer domesticus* showing multinucleated giant cell, 10 × 70.

No hypertypic giant cells could be seen as reported in mammals by other workers[4,5]. Most of the multinucleated giant cells were derived from the spermatocytes asd rarely from the young spermatids. The mode of formation of these cells as observed during the present investigations was due to the fusion of the cleaving cells. Rao and Srivastava[5], had suggested that giant cells could be formed by cell fusion, possibly by fatty degeneration of cell membrane. Montgomery *et al.*[6], had also reported that giant cells were formed by the fusion of the plasma membrane.

According to Amoroso[7], the giant cells were generally formed because of the macrophages which swell the young developing spermatids.

The present observations indicate the lag effect due to irradiation thus interfering with the regular spermatocytic divisions and thereby causing the fusion instead of forming the spermatids. The detailed chemical sequence will be published eslewhere.

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---

1. Barratt, J. O. and Arnold, G., *Arch Zellforsch*, 1912, 7, 242.
2. Pitcock, J. A., In : *Effects of Ionising Radiation on the Reproductive System*, Ed., W. D. C. Arlson, and F. X. Gassner, Pergamon Press, London, 1964, p. 156.
3. Deschner, E. E., Rugh, R. and Grupp, E., *Milit. Med.*, 1960, 125, 447.
4. Bhatia, A. L., *Curr. Sci.*, 1975, 44, 470.
5. Rao, A. R. and Srivastava, P. N., *Experimentia*, 1967, 23, 381.
6. Montgomery, P. O. B., Karney, D., Reynolds, R. C. and Mcclendon, D., *Am. J. Path.*, 1964, 44, 727.
7. Amoroso, E. C., In : *Radiation Effects in Physics. Chemistry and Biology*, Ed. M. Ebert and A. Howard, North-Holland, Publishing Co., Amsterdam, 1963, p. 424.

CYTOLOGICAL STUDIES IN ATEMOYA (*ANNONA ATEMOYA* HORT.)

ATEMOYA is a hybrid between Cherimoya (*A. cherimola* Mill.) and Custard apple (*A. squamosa* L.). Atemoya is a distinct improvement in respect of seedlessness and fruit quality on either of the parents. The low bearing habit of this crop is considered to be its greatest drawback. The studies on cytological aspects, *viz.*, pollen sterility, pollen germination and meiosis were undertaken to know the cytological reasons associated with low fruit set.

The pollen sterility, in the present study, was high (Table I), during May and June when the temperature was high and humidity low. Thakur and Singh[1] observed that the pollens were not fertile till the end of July.

TABLE I
Pollen sterility in Annona atemoya

Month	% of sterile pollens	Monthly Av. Temp. in °C Max.	Min.	Av. Relative Humidity %
May	58	39	17	38
June	55	37	15	36
July	33	34	14	66
August	30	31	13	74

The pollen grain germination was tried in sucrose solution (10, 15, 20 and 25%), *plus* agar-agar (1%), stigmatic secretion of *A. atemoya, A. squamosa* and *A. atemoya plus A. squamosa,* Gibberellic acid (10, 25, 50 and 75 ppm) and water. The maximum pollen germination (8·6%) was observed in stigmatic secretion of *A. atemoya.* The low pollen germination may be due to the compound nature of pollen grain with thick exine. Thakur and Singh[1], reported 11·6% pollen germination in 20% sucrose solution.

The flower buds for meiotic studies were fixed in propiono-acetic alcohol and squashed in 1% propiono-carmine. The meiotic study confirms $n = 7$ as chromosome number of Atemoya. The regular meiosis was observed in only 52% of the cells (Table II). The presence of univalents was due

TABLE II
Chromosomal associations at Diakinesis and Metaphase in Annona atemoya

No. of PMC's observed	Chromosomal bodies	Univalents	Bivalents	% of particular association
4	9	4	5	16
3	8	2	6	12
1	13	12	1	4
2	14	14	0	8
1	10	6	4	4
13	7	0	7	52
1	11	8	3	4

to the lack of pairing and the most common configuration was found to be 5 II + 4 I. However, data reveal irregular meiosis in 48% cells. Thakur and Singh[2], reported $n = 7$ as chromosome number of Atemoya and observed varying number of univalents in some of the cells.

The chromosomal separation in anaphase-I (Table III) was abnormal in 36% of the cells. Three chromosomal groups were also observed in some of the cells. This extra group of chromosomes was due to the formation of laggards which might have resulted from the failure of chromosomal pairing. Thakur and Singh[2], have also reported unequal anaphase separation and three groups of chromosomes.

TABLE III

Distribution of chromosomes at anaphase-I in Annona stemoya

Sl. No.	Extent of un/equal distribution	No. of cells	% of cells
1.	PMC's with equal distribution as 7:7	16	64
2.	PMC's with unequal distribution as (1) 6:8	1	4
	(2) 5:9	1	4
3.	PMC's with unequal distribution with laggards	7	28

The present study indicated high (43·9%) pollen sterility which is due to irregular meiosis (48%) and subsequent abnormal chromosomal distribution (36%). The low pollen fertility and germination finally hinder fruit setting in *A. atemoya*.

Marathwada Agricultural
University,
Parahani,
August 28, 1975.

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S. T. BORIKAR.
N. N. SHINDE.
U. G. KULKARNI.

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1. Thakur, D. R. and Singh, R. N., *Ind. J. Hort.*, 1965, 22, 10.
2. — and —, *Ind. J. Genet.*, 1965, p. 367.

OLIGOGYNOPHORIA, A VIRUS INDUCED LESION IN THE GROUNDNUT, *ARACHIS HYPOGAEA* L.

FOLLOWING the report of the new virus, infecting the groundnut *Arachis hypogaea* L.[1], physiological alterations brought about by this virus on its host were studied. These studies[2] and field observations formed the basis for postulation of "oligogynophoria" as a pathological condition resulting in the host plant under the hegemony of groundnut chlorotic spot virus. The term "oligogynophoria"[2] was tentatively defined as "pathological reduction in the number of gynophores".

With an aim to obtain a clearer definition of oligogynophoria in the groundnut plant under viral influence, a batch each of healthy and infected (under apparent attack of groundnut chlorotic spot virus) plants were collected from fields near Nagari, Andhra Pradesh. A visual examination of the healthy and infected "fruit systems" revealed *prima facie* oligogynophoria (Fig. 1). Three types of gynophores were marked out for statistical processing[3] of numerical data. Fruiting gynophores : gynophores with terminal fruits with hardened shells ; headed gynophores : with terminal fruits in which no hardening of shell was noticeable[3] ; headless gynophores : bearing no terminal fruit (gynoecium) at all. The total gynophores (aggregate of fruiting, headed and headless gynophores of each fruit system) showed no statistically significant difference between healthy and affected plants (Table I). Viral attack evidently does not cause numerical diminution of gynophores, Even the headless gynophores (which probably represent flowers which were not cleistogamously fertilized) exhibited no statistical alteration under viral infection. On the other hand the "fruiting" and "headed" gynophores showed statistically significant alterations under viral influence. The numbers of fruiting and headed gynophores exhibited an inverse alteration under viral affectation : The fruiting gynophores were predominant in the healthy plant over headed gynophores and in the virus-affected plants, the "headed" gynophores were preponderant. In the healthy (virus-unaffected) plants greater number of headed gynophores should obviously have been driven into the condition of "fruiting" ;

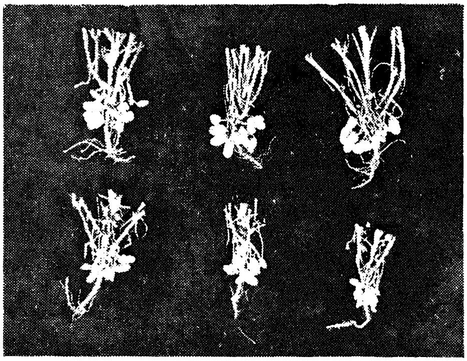

FIG. 1. Oligogynophoria in groundnut (*Arachis hypogaea* L.) under the influence of groundnut chlorotic spot virus. Upper row—Healthy "fruit systems" ; Lower row—Virus-affected "fruit systems".

TABLE I

Effect of groundnut chlorotic spot virus infection on the fruit system of the host, Arachis hypogaea *L.*

Parameter	Healthy	Virus-affected	*b*	*c*	*d*	*e*
Fruited gynophores	*a* 11·55 ± 2·32 (9)	2·0 ± 1·142 (10)	10·97	17	$P < 0{\cdot}001$	− 82·68
Headed gynophores	4·78 ± 4·15 (9)	10·3 ± 4·58 (10)	2·741	17	$P < 0{\cdot}02$	+115·50
Headless gynophores	8·22 ± 6·85 (9)	7·5 ± 4·03 (10)	0·283	17	N.S.	..
Total gynophores	25·56 ± 10·96 (9)	19·8 ± 7·34 (10)	1·123	17	N.S.	..
Fruiting gynophores per cent total gynophores	56·10 ± 26·31 (9)	11·95 ± 11·09 (10)	4·684	17	$P < 0{\cdot}001$	− 78·71

a: Values are means ± standard deviations; The numbers of observations are parenthesized.
b: calculated students '*t*' test value.
c: number of degrees of freedom.
d: level of significance: $P < 0{\cdot}02$: significant; $P < 0{\cdot}001$: highly significant; N.S.: not significant.
e: change in affected plant per cent healthy plant.

under viral influence, the process of "fruiting" of headed gynophores seems to be greatly retarded. Hence the oligogynophoria can be redefined as "pathological reduction of the number of fruiting gynophores or pathological retardation of the process of fruiting resulting in a smaller of headed gynophores reaching the stage of fruiting". The authors were informed by the farmers during the collection of the plants for the present study, that the "fruiting" gynophores alone persist as fruit. Oligogynophoria can therefore be taken as a well founded field lesion with considerably profound implication on yield economics of groundnut crop.

The studies on physiological alterations under the chlorotic spot viral influence in the groundnut plant[2] were confined to TMV-2 which is locally, a preferred variety for cultivation practices. But the infective action of this virus extends beyond this variety to many other groundnut varieties (TMV$_3$, HG$_8$, HG$_{10}$, Aceria mutand) and many more papilionaceous hosts also.

The virus has been shown[2] to act on its groundnut host and in other susceptible hosts[4], in the cotyledonotrophic stage of the growing seedling. It has also been indicated that the action of virus is directed at the metabolism of γ-methylene glutamine[2], the principal translocative source of nitrogen[5] in the "cotyledonotrophic" stage of groundnut plant. The pathological diversion of "nitrogen" available from this principal translocative source into the viral biosynthetic pathway has been indicated[2] to cause a "famine of nitrogen" exactly at a stage when gynophorogenesis occurs. This famine might be the decisive factor in bringing about retardation of the process of fruiting and resultant oligogynophoriac lesion. Added to this lesion, the number of root nodules occurring on the root system of the host plant also seems to be affected[6] leading to a further restriction of nitrogen supply and poorer increase in size of the "fruited" gynophores.

We are thankful to Prof. V. S. Rama Das, for encouragement and facilities.

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1. Haragopal, T. and Nayudu, M. V., *Phytopath. Z.*, 1971, 71, 33.
2. —, *Doctoral Thesis Submitted to Sri Venkateswara University*, Tirupati, 1975.
3. Pillai, S. K. and Sinha, H. C., *Statistical Methods for Biological Workers*, Ramprasad and Sons, Agra-3, India, 1968.
4. Haragopal, T. and Chandrasekharam, V., In Preparation, 1975 *a*.
5. Fowden, L., *Ann. Bot.*, 1954, **18**, 417.
6. Haragopal, T. and Chandrasekharam, V., In Preparation, 1975 *c*.

LEAF SURFACE IMPRINTING—A NEW, EASY AND CHEAP METHOD

THE present day methods to take the imprints involve the use of mucilage from unripe and ripe fruits[2-4], domestic adhesives[1] and silicon rubber plastic[3]. In the present study a new and easy device for making epidermal imprints by using domestic 'Maida' is described. Maida is a fine wheat flour mainly starchy in nature when tested with iodine. Commercially available 'maida powder' was mixed with adequate amount of distilled water and boiled and continuously stirred so as to make it a semi-liquid paste. The paste was allowed to cool. The paste was then applied uniformly on the surface of the leaf with the help of a camel hair brush. The leaf, on the surface of which the paste has to be applied need not be plucked from the plant, and the surface was cleaned to remove exotic particles with a moist fine clean muslin cloth. The paste applied leaves were kept in the open air on the plant itself for about 10–15 minutes to dry. Drying will depend upon the humidity, wind and temperature also. After the paste has dried up, it was stripped off with utmost care using fine forceps and needles. The film was then mounted with the imprinted surface uppermost on a dry clean glass slide. The coverslip was gently tapped to make the film uinformly flat. Repeated imprints were taken from the same surface of the leaf of a plant without causing any injury either to the leaf or plant. Plants from different groups were tested. The imprints of the leaf surface were examined with normal light microscope using direct illumination. Microphotographs presented were taken with ORWO DK 5 film using yellow filter and incandescent light. The microphotographs (Fig. 1) reveal the satisfactory results of this new method. The method holds good for herbarium specimens also. This method will be useful not only to plant morphologists but also to physiologists as repeated imprints can be taken from the same surface of the leaf of a plant at intervals.

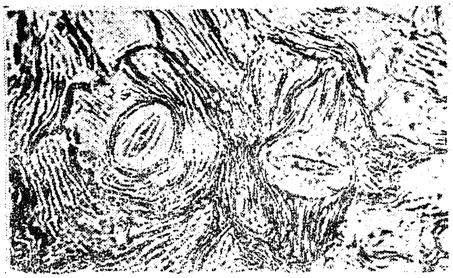

FIG. 1. *Thespesia populnea* Soland., : Imprint of leaf-abaxial surface. Note cuticular striations and stomata, × 880.

One of us (M. Gangadhara) thanks the U.G.C. for the award of a junior research fellowship.

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1. Inamdar, J. A. and Patel, R. C., *J. Microscopy*, 1969, 90, 269.
2. —, — and Bhatt, D. C., *Zt. Wiss. Mikr. Mickrosc. Technik*, 1970, 70, 140.
3. Janny Groot, *Acta Bot. Neerl.*, 1969, 18, 703.
4. Patel, R. J., *Curr. Sci.*, 1968, 37, 680.

EXILISPORA INDICA SP. NOV. (SPHAERIACEAE)—A NEW GENERIC RECORD TO INDIA

A RARE ascomycetous fungus was collected on the dead stems of *Gymnosporia rothiana* Mont. (Fam. Celastraceae) in the Forests of Bhimashankar, 100 Km away from Poona, in the month of September, 1974. On examination this proved to be an undescribed species of the genus *Exilispora* Tehon and Daniels[2] (Sphaeriaceae). This genus is monotypic and remained unrepresented in the fungal flora of India so far.

Exilispora indica sp. nov. (Fig. 1).

Perithecia nigra, solitaria, sub-erumpentia, septata, ostiolo-rostrata. periphysata, magnit 172–280 × 184–260 µ ; rostra 1·5 mm longa ; asci cylindracei vel clavati, brevi-stipitati, octospori, aparaphysati, unitunicati, 57–112 × 5·4–9 µ ; ascosporae sub-hyalinae vel olivaceae, scoleciformae, septatae (9–12), magnit 52–72 × 3–5 µ.

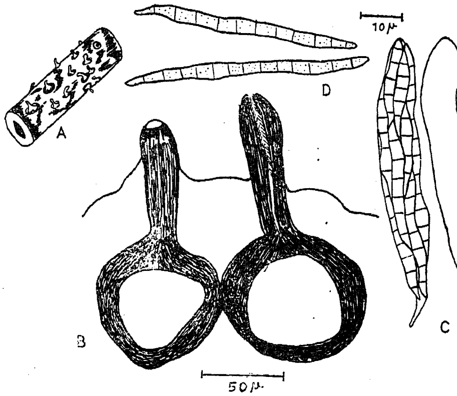

FIG. 1. *Exilispora indica* sp. nov. A, Habit ; B, Perithecia (V. S.); C, Asci ; D, Ascospores.

Matrix : In caules *Gymnosporia rothiana* Mont. Legit. A.W.S. ad Bhimashankar (30–9–1974) ; No. AMH 2667 (Holotypus).

The present collection differed significantly from the type species, *viz., Exilispora plurisepta* Teh. and Dan.[2], in gross-morphology, dimensions of perithecia, asci and ascospores besides being collected on a new host. Although the genus *Exilispora* closely resembles the genus *Ophiobolus* Reiss.[1], it differs from it in having significantly long beaked perithecia and phaeo-scolecosporus ascospores.

Thanks are offered to Prof. M. N. Kamat, for his keen interest, to the Director, for facilities, to Dr. Mrs. A. Pande, for helpful suggestions and to Government of India, for the award of the S.R.F. to the senior author.

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September 15, 1975.

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1. Ainsworth, G. C., Sparrow, F. K. and Sussman, A. S., *The Fungi,* Academic Press, New York and London, 1973, **4 A**, 621.
2. Tehon, L. R. and Daniels, E. Y., *Mycologia,* 1927, **19**, 110.

VIVIPARY IN *LIVISTONA CHINENSIS* R. Br.

IN Monocotyledons, habitual vivipary has so far been described only in *Pectinella antarctica* Black, a marine member of Potamojetonaceae. It also occurs as a rarity in *Meloccana bambusoides* of Graminae. Since there are no published reports of vivipary in palms an account of viviparous seedlings in *Livistona chinensis* is given here.

On 22nd July 1975, a number of fruits of *L. chinensis* in germinated stage were noticed while they were still attached to the fruitiferous axis (Fig. 1). In these fruits, radicular portion of the embryo was peeping out through the fleshy green epicarp. The fruitiferous axis was sprayed with an insecticide to avoid ant attack and was loosely covered with moist thin muslin cloth to provide sufficient moisture. Observations were made until 10th August and the results noted.

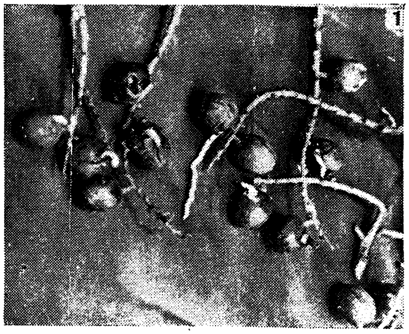

FIG. 1

Nearly 79% fruits got abscissed when the seedlings were ranging from 1 to 1·5 cm in length. Only in exceptional cases the seedlings could grow upto 4·5 cm when the fruits were attached to the tree (Fig. 2). In none of the seedlings the plumule could be seen emerging from the cotyledonary sheath. The radicle also did not elongate much.

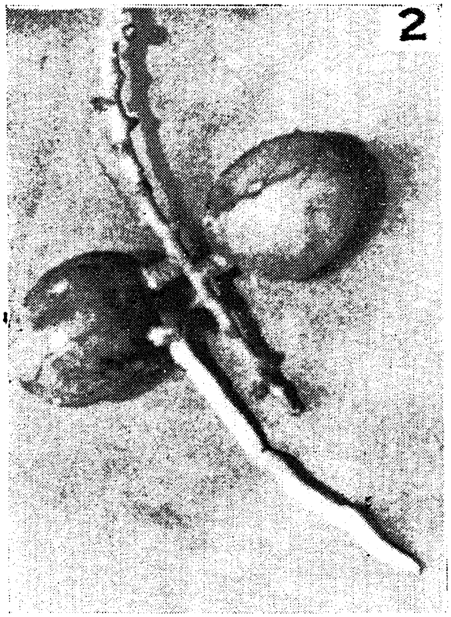

FIG. 2

In the normal course, the mature fruits of *Livistona chinensis* get abscissed when the epicarp turns to deep black colour. In a few cases they remain attached until the pericarp completely dries up. The mature fresh fruits when placed in soil germinate only when the pericarp is partially or completely decayed. This process takes two to six weeks depending upon the season. The relative humidity during these days was ranging from 73 to 98%. The precoutius germination of fruits reported here might be due to high humidity of the atmosphere.

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Ramnarain Ruia SHASHI BHUSHAN PANDEY.
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Bombay 400 019, *September* 12, 1975.

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1. Staf, O., "On the fruit of *Melocanna bambusoids,* Trino, an endospermless, viviparous genus of Babmbuseae," *Trans. Linn. Soc. Lond. Bot.*, Ser. II, 1904, **6**, 401.

SOME QUANTITATIVE OBSERVATIONS ON THE BRAIN OF THE CAT-FISH, *HETEROPNUESTES FOSSILIS* (BLOCH.)

THOUGH considerable literature has accumulated on the anatomy of fish brain[1], the information on the morphometric and allometric aspects has been rather limited[2-4]. The present investigation was undertaken with a view to analysing the intra-specific relationships between the length/weight of the brain, brain/body lengths and brain/body weights of *Heteropnuestes fossilis* (Bloch.), a common freshwater cat-fish. Such quantitative study would be useful in analysing the absolute and relative growth characteristics of teleostean brain as also in working out the degree of interspecific variations in the established relationships.

Fishes of the size range 125 to 205 mm were obtained from the fish market. After recording the total body length and weight, the brain of each specimen was taken out, weighed and its length accurately measured. In all, 72 fishes were examined. The various relationships were expressed through regression equations, using methods of calculations as given by Snedecor[5].

Growth in the body length of the fish was found to be accompanied by an increase in the length of the brain (Fig. 1 A). This was found to be a constant feature in fishes of all the size ranges investigated. This relationship was expressed by the equation :

$$\text{Log } x_1 = 0\cdot9548 + 0\cdot9944 \log y_1$$

Where, x_1 was the body length of the fish (mm), and y_1 was the length of the brain (mm).

The correlation coefficient (r_1) for this relationship was found to be $0\cdot298 \pm 0\cdot344$, significant at 10% level ($P > 0\cdot10$), $Z = 0\cdot3$.

Further, increase in the body weight of the fish seemed to maintain a straightline relationship with the increase in the weight of the brain (Fig. 1 B). This relationship could be expressed by the following logarithmic equation :

$$\text{Log } x_2 = 0\cdot0207 + 1\cdot5469 \log y_2$$

where, x_2 was the body weight of the fish (mg), and y_2 was the weight of the brain (mg).

The coefficient of correlation, r_2 ($0\cdot670 \pm 0\cdot209$) was significant at 1% level ($P > 0\cdot01$), $Z = 0\cdot8$.

The regression equation establishing the length/weight relationship of the brain was computed to be

$$\text{Log } x_3 = 0\cdot0856 + 0\cdot4765 \log y_3$$

where, x_3 was the length of the brain (mm), and y_3 was the weight of the brain (mg).

The above relationship has been shown in Fig. 1 C. The correlation coefficient (r_3) for the same was calculated as $0\cdot886 \pm 0\cdot080$. Using the distribution of 't' as a measure of probability, r_3 was found to be significant at 1% level ($P > 0\cdot01$), $Z = 1\cdot4$.

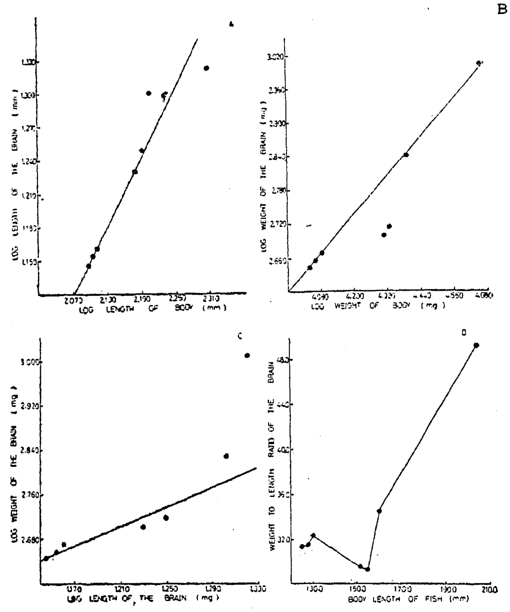

FIG. 1

The growth in the weight of the brain, however, did not proceed uniformly with increase in its length. This was evident from the variations in the weight to length ratio of the brain (Fig. 1 D). This ratio was found to be specific for the different size-groups of *H. fossilis*. In the fishes for the sizes investigated the ratio seemed to rise steadily from 31·4 to 32·4 as the body length increased from 125 to 130 mm. Further increase in the size of the fish was characterized by a decline in this ratio to 29·2 in the specimens of 155 mm size-group. In the larger size-groups, however, the weight of length ratio was again found to rise, indicating that the growth in the weight of the brain continued to occur even when the growth in the brain length, and so also the body length, considerably slowed down.

We are greatly indebted to Dr. M. H. Kasim, Head, Department of Biology, College of Science, University of Mosu, Mosul, for providing the laboratory facilities and for his keen interest in the progress of the work.

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1. Bernstein, J. J., In: *Fish Physiology* (Hoar, W. S. and Randall, D. J., Eds.), Academic Press, London and New York, 1970.
2. Bauchot, R., Platel, R., Ridet, J. and Thireau, M., *Acta Zoologica,* 1973, **54**, 53.
3. Geiger, W. Von, *Acta Anat.,* 1956, **26**, 121.
4. —, *Ibid.,* 1956, **27**, 324.
5. Snedecov, G. W., *Statistical Methods,* The Iowa State College Press, Iowa, 1955.

ULTRASTRUCTURE OF FLAGELLA OF SPERMATOZOIDS OF *LYGODIUM FLEXUOSUM* (L.) SW.

FINE structure of the flagella of spermatozoids of various plants and plant groups, *viz.*, *Stigeoclonium*[4], *Polytrichum*[7], *Marchantia*[1,2], *Marsilea*[8], and *Zamia*[5,6] has been investigated in detail. Our investigations on the ultrastructure of the flagella of spermatozoids of *L. flexuosum* have brought out certain new facts in the flagellar construction of the fern spermatozoids which are reported here.

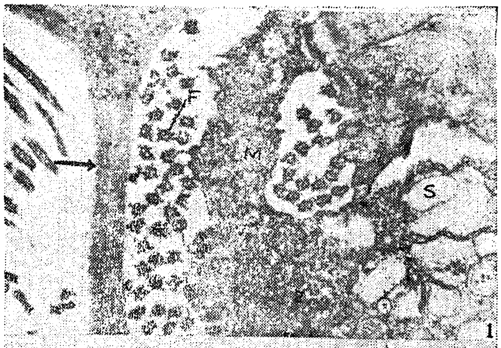
Spores of *L. flexuosum* were collected from the garden of Botany Department, Lucknow University, and were sown under sterile conditions in 1% Knop's solution. The temperature of the culture remained at 24 ± 2° C with light intensity at 600 ft. c.

Spores, germinated on the 4th or the 5th day, though antheridia began to develop after 70 to 80 days. Gametophytes with mature antheridia ready to discharge spermatozoids were fixed in Caulfield's[3] fixative at 4° C for 4 hours. After dehydration, the prothalii were embeded in plastic mixture of Epon and Araldite and ultrathin section were cut on LKB ultratome III, using glass knives. Silver grey colour sections were picked on uncoated copper grids. Sections were stained in lead citrate and uranyl acetate and examined under Hitachi electron microscope Hu-11 E at 75 kV. Photographs were taken on Fuiji electron microscopic film.

The micrographs of the spermatozoids pass through the region where flagella are attached to Vierergruppe. A giant mitochondrion (M) having numerous cristae in very close association of the Vierergrappe as well as numerous flegella cut transversely (Fig. 1) can also be seen in cross-section.

Interesting features of the flegella as seen in the cross-section of the median portion is that the outer membrane has sharp projections and is star shaped (Fig. 2). This differs from the findings of earlier workers who noticed smooth outer membrane for the flagella. However, basal part of the flagellum has smooth outer wall (Fig. 2 shown by arrow).

We thank Dr. Nitya Nand, Director and Dr. S. C. Agarwal, Head of the Department of Biophysics. CDRI, Lucknow, for permission to use electron microscope. We also thank Sri. A. C. Shipstone, Dr. S. C. Maitra and Sri. V. K. Bajpai of the same institution for their active and willing help in the use of electron microscope.



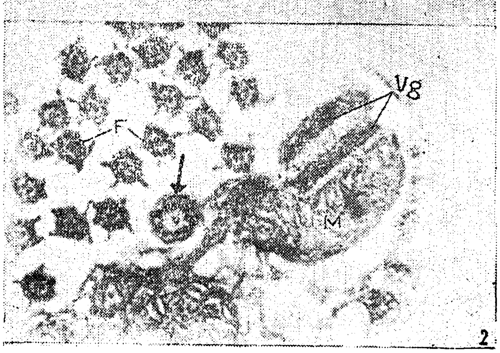


FIG. 1. T.S. through antheridia showing:

F.—Star-shaped flagella, M.—Mitochondrion. S.—Starch, Arrow showing cell wall (× 15,000).

FIG. 2. T. S. through antheridium showing:

F.—star shaped flagella, M.—Mitochondrion, Vg.—Vierergruppe. Arrow showing T.S. Basal part of flagellum (× 30,000).

The junior author is grateful to UGC for providing financial assistance for this project.

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USHA KHER.

1. Carothers, Z. B. and Kreitner, G. L., *J. Cell Bio.*, 1967, **33**, 43.
2. — and —, *Ibid.*, 1968, **36**, 603.
3. Caulfield, J. B., *J. Biophys. Biochem. Cytol.*, 1957, **3**, 827.
4. Manton, I., *J. Exptl. Botany*, 1964 *a*, **15**, 399.
5. Norstog, K., *Amer. J. Bot.*, 1967, **54**, 831.
6. —, *Ibid.*, 1974, **61** (5), 449.
7. Paolillo, D. J. Jr., *Can. J. Bot.*, 1965, **43**, 669.
8. Rice and Lastsch., *Amer. J. Bot.*, 1967, **54**, 856.

AN ADDITION TO INDIAN HYPHOMYCETES

DURING taxonomic studies on the microfungi of Gorakhpur (U.P.), a rare hyphomycete was isolated in July, 1975 from a petridish containing malt extract agar medium which was exposed to air. The fungus was first described as a new species of *Chloridium*, *C. apiculatum*, by Miller, Giddens and Foster[1] in 1957 from forest soil of Georgia, U.S.A. As the sporogenous cell of this fungus is not a phialide, the species cannot be retained in the genus *Chloridium*. Dr. M. B. Ellis of the Commonwealth Mycological Institute, Kew, has informed the authors in a personal communication that he has transferred the fungus to the genus *Veronaea*, making a new combination *Veronaea apiculata* (Miller, Giddens and Foster) M. B. Ellis, in his forthcoming book *More Dematiaeeous Hyphomycetes* (In press). The fungus is reported here for the first time from India. A brief description of the Indian isolate is given below.

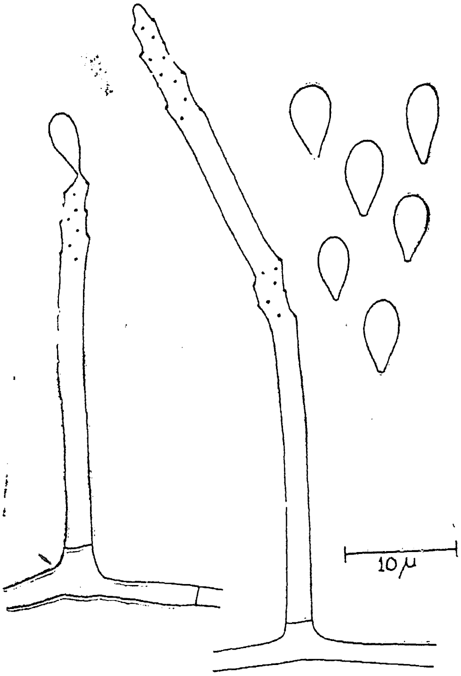

FIG. 1. Conidiophores and Conidia.

Colonies on malt extract agar up to 2 cm in diam. in 10 days, grey in colour. Mycelium superficial and submerged. Conidiophores arising from aerial hyphae, macronematous, mononematous, simple, straight or flexuous, with a septum near the base, light brown, 24–55 μ long, 1·5–2·2 μ in diagm., cicatrized at the apex; often groups of conidial scars along the length of the conidiophore alternate with smooth areas. Conidia obovoid or pyridorm, with narrow truncate base and rounded apex, subhyaline, smooth 0-septate, 4·4–6·6 × 2·0–3·3 μ (Fig. 1).

Isolated from air, Gorakhpur (U.P.), India, July 1975. A dry petri dish culture has been deposited in the Commonwealth Mycological Institite, Kew, as IMI 195484.

The authors wish to thank Dr. M. B. Ellis for his help in the identification of this fungus.

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Gorakhpur 273001, U.P., K. S. BHARGAVA.
September 16, 1975.

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1. Miller, J. H., Gidden. J. E. and Foster, A. A,, *Mycologia*, 1957, **49**, 779.

THE ROOT-KNOT NEMATODE ON COMMERCIAL PLANTINGS OF TUBEROSE IN INDIA

DURING a survey of some of the tuberose *Polianthes tuberosa* Linn. plantations in Bangalore District for plant parasitic nematode population, it was observed that root-knot nematodes were commonly encountered causing heavy root-galling. The plants were stunted with chlorotic foliage and short and thin floral stalks. The nematode species *Meloidogyne incognita* (Kofoid and White, 1919). Chitwood (1949), was found to be associated with severe galling of the roots. The offsets were not found to be infested. Both the types of tuberose, *viz.*, "Single" and "Double" under cultivation were found to be equally susceptible to *M. incognita* at the Eeperimental Station, Indian Institute of Horticultural Research, Hessaraghatta, Bangalore,

Root-knot nematodes on container grown tuberose had been reported from U.S.A.[1] and Israel[2]. This is the first report of root-knot nematode damage to commercial tuberose plantations from India. As the nematode is found on the roots of tubers, thorough removal of roots at the time of harvest and storage of tubers will check the spread of these nematodes.

Thanks are due to Dr. S. S. Negi, Senior Geneticist (Floriculture) and to Dr. G. S. Randhawa, Director, for their keen interest in these studies.

Indian Institute of Horticultural V. R. RAO.
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255, Upper Palace Orchards,
Bangalore-560006, India,
January 14, 1976.

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1. Johnson, A. W., *J. Nematol.*, 1970, **2**, 191.
2. Minz, G., *Spec. Bull., Agr. Res. Stn., Rehovot*, 1958, **12**, 10.

SHORT SCIENTIFIC NOTES

Isolation of Carpesterol from the Fruits of *Solanum torvum*

Solanum torvum (Family: Solanaceae), a useful Indian medicinal plant has been earlier studied for its chemical constituents[1-3]. The present work records the isolation of carpesterol for the first time from this plant.

Shade dried fruits were extracted with hot 80% ethanol under reflux. The ethanol concentrate was repeatedly shaken with benzene, ether and ethyl acetate. The benzene soluble portion on elaborate column chromatography over neutral alumina yielded in benzene : pet. ether (3 : 1) eluate, a pure compound (0·05% yield), m.p. 245–7°, $[\alpha]_D + 61 \cdot 5^\circ$ (chf) and M.F. $C_{37}H_{54}O_4$. It gave positive Liebermann–Burchard and Salkowski tests and formed an acetate, m.p. 187–9°. This sterol was identified as carpesterol by direct comparison with an authentic sample by m.m.p., co-TLC (sillca gel impregnated with $AgNO_3$) and The other fractions yielded a low melting solid (m.p. 62–4°), and β-sitosterol (m.p. 131–3°).

Though carpesterol was isolated, four decades ago from *S. xanthocarpum*[4], its structure was only recently elucidated[5]. The present isolation of this sterol from *S. torvum* as a second source may be of some chemotaxonomic interest.

We thank Dr. J. A. Beisler, National Cancer Institute, Bethesda, U.S.A., for the kind supply of an authentic sample of carpesterol, Prof. M. N. Ghosh, and the Principal, JIPMER, for encouragement.

Departments of Pharmacology and Chemistry,
Jipmer, Pondicherry 605 006,
November 19, 1975.

T. K. Bhattacharya.
T. N. C. Vedantham.
S. Sankara Subramanian.

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1. Krishnamurti, G. V. and Seshadri, T. R., *J. Sci. Ind. Res. (India)*, 1949, 8 B, 97.
2. Chopra, R. N., Chopra, I. C. and Varma, B. S., *Supplement to Glossary of Indian Medicinal Plants*, Publications and Information Directorate, New Delhi, 1969, p. 93.
3. Mythirayee, C., Krishnamurty, V. and Madhavakrishna, W., *Curr. Sci.*, 1975, **44**, 461 and references cited therein.
4. Saiyed, I. Z. and Kanga, D. D., *Proc. Indian Acad. Sci.*, 1936, **4 A**, 255.
5. Tsay. Y. H., Silverton, J. V., Beisler, J. A. and Sato, Y., *J. Amer. Chem. Soc.*, 1970, **92**, 7005; Beisler, J. A. and Sato, Y., *J. Org. Chem.*, 1971, **36**, 3946.

Note added in proof: Since communicating our work on *S. torvum*, carpesterol has also been isolated by us from the fruits of *S. trilobatum*.

Occurrence of *Macrognathus aculeatus* (Bloch) in the River Cauvery at Mekedatu, Karnataka State

Two Specimens of the Lesser Spiny Eel, *Macrognathus aculeatus* (Bloch) were collected on August 18, 1974, while operating a cast net in the rock-strewn stretch of the river Cauvery at Mekedatu about 60 Km from Bangalore. Identification of the species was confirmed by the presence of 5 ocelli like spots on the dorsal fin. The present species being rare, has not been reported so far from the upper reaches of Cauvery river above Hoginakal falls and in this paper the details of the two specimens are reported.

Of the two specimens collected, one is a juvenile with T.L., 150 mm and 3·5 g in weight and the other an adult female with T.L., 287 mm and 24·5 g in weight. The examination of the gonad of the adult specimen revealed that the fish was in the first stage of maturity. The juvenile specimen of *M. aculeatus* collected from Cauvery river may belong to 1 + age group[2]. The river Cauvery is rock-strewn and shows seven individual falls at Hoginakal and this falls become flush with the rise of about 30 meters in the level of the Mettur reservoir (India), the fish like *Catla catla* and *Pangasius pangasius* can ascend up the river[1]. The occurrence of *M. aculeatus* in the river Cauvery, about 400 Km from the deltaic region strongly suggests that it is capable of ascending very long distances in the river negotiating many water falls, especially during juvenile stages similar to the up-stream migration of elvers of *Anguilla nebulosa labiata* which ascend Tana River in Kenya, overcoming many water falls including a vertical fall of 23 meters[3].

Our thanks are due to Dr. A. R. Kashuri Bai, for her encouragement.

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University of Agricultural Sciences,
Bangalore 560 024, *October* 10, 1975.

H. R. Srinivasachar.
K. V. Rajagopal.
Dwaraka S. Rao.

---

1. David, A., Govind, B. V., Rao, N. G. S. and Rajagopal, K. V., *Indian J. Fish.*, 1967, **14**, 54 (Issued 1971).
2. —, Ray, P., Govind, B. V., Rajgopal, K. V. and Banerjee, R. K., *Bull. Cent. Inl. Fish. Res. Inst. Barrackpore*, 1969, No. 13, 83.
3. Frost, W. F., *Colonial Office*, East Africa, 1954, Fishery Publication No. 6.

Additional Host Records of the Root-Knot Nematode, *Meloidogyne incognita* in North India

It is well known that the root-knot nematodes belonging to the genus *Meloidogyne* cause enormous loss to many economically important crops. They have a very wide host range in both cultivated and wild plants. Despite this there might be many more hosts which await the report. Hence, a survey was done in the Aligarh District (North India) for host range and host susceptibility of the prevailing species of the nematode. The specific identification was done by close examination of the perineal patterns of mature females, taken out from galled tissues of roots. Hot acid fuchsin (0·01%) and lactophenol were used as staining and mounting materials respectively. The species attacking all the plants, reported here, was found to be *Meloidogyne incognita* (Kofoid and White 1919) Chitwood, 1949. The root-knot infection was rated as + = light (1–25%), ++ = moderate (26–50%), +++ = heavy (51–75%) and ++++ = severe (76–100%) on the basis of percentage of roots infected as suggested by Smith and Taylor[10]; and the size of root galls recorded as small (S), medium (M) and large, (L), All the plants listed below are new host records of *M. incognita* which have not been reported earlier[1–9,11]:

New host records: *Acacia arabica* Willd. (+ ; S), *A. farnesiana* Wall. (+; S), *Calotropis procera* Bry. (++; S, M), *Cassia occidentalis* L. (+; S), *Centaurea cyanus* L. (++ : M, S), *Fumaria parviflora* Lamk. (+; S), *Hibiscus rosa-sinensis* L. (++; S) *Ipomoea carnea* Facq. (++; M), *I. eriocarpa* R. Br. Prod. (+++; M), *Launaea asplenifolia* Hook. (++; S, M), *Portulaca quadrifida* L. (++; M, S), *Rumex hastatus* Don Prod. (+; S), *Sida cordifolia* L. (++; S, M), *Spinacia oleracea* L. (++++; L, M), *Vicia hirsuta* (L.) Gray. (+++; M).

First report from India: *Antirrhinum majus* L. (++++; L), *Cichorium intybus* L. (+; S).

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Aligarh–202001, India, *September* 26, 1975.

M. Mashkoor Alam.
Abrar M. Khan.
S. K. Saxena.

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1. Alam, M. M., *Curr. Sci.*, 1975, **44**$_1$ 445.
2. —, Khan, A. M. and Saxena, S. K., *Indian J. Nematol.*, 1973, **2**$_1$ 159.
3. Bhatti, D. S., Gupta, D. C., Dahiya, R. S. and Malhan, I., *Curr. Sci.*, 1974, **43**$_1$ 622.
4. Davidson, T. R. and Townshend, J. L., *Nematologica*, 1967, **13**, 452.
5. Goodey, J. B., Franklin, M. T. and Hooper, D. J., *The Nematode Parasites of Plants Catalogued Under Their Hosts* (3rd Ed.), C.A.B., England, 1965.
6. Nigam, B. C. and Reddy, A. S., *Indian Phytopath.*, 1972, **25**$_1$ 315.
7. Potter, J. W., Townshend, J. L. and Davidson, T. R., *Nematologica*, 1969, **15**, 29.
8. Roy, A. K., *Indian J. Nematol.*, 1972, **2**, 86.
9. Sitaramaiah, K., Singh, R. S., Singh, K. P. and Sikora, R. A., *Plant Parasitic and Soil Nematodes of India*, Exp. Sta. Bull. No. 3, U.P. Agr. Univ., Pantnagar, 1971.
10. Smith, A. L. and Taylor, A. L., *Phytopathology* 1947, **37**, 85.
11. Whitehead, A. G., *Nematologica*, 1969, **15**, 315.

Neoascaris vitulorum Larvae in the Colostrum/Milk of Buffaloes

Neoascaris vitulorum infection was found in 26% (64 out of 224) buffalo calves below the age of 6 months on the basis of their faecal examination. Some workers have recently reported that the passage of infective larvae of certain nematode parasites through the colostrum/milk of infected dams forms an important and in some cases a major source of infection for their new-born calves[1–4].

One hundred and eighty-seven milk samples (250 ml. each) were collected for examination from 37 buffaloes, 1–30 days after parturition. The milk was collected from all the four quarters after thoroughly washing the udder and the teats. Four buffaloes, out of 37 were found positive for the presence of *N. vitulorum* larvae in their milk from 1–11 days after parturition. The larvae resembled those of *N. vitulorum*, except that they were twice the size of the normal infective larvae which is in agreement with the observations of Warren (1969)[3]. They were in an advanced stage of development, as evidenced by their morphological structures, which may explain their early maturity to adult worms in young calves, as early as 7 days after birth[5].

The faecal examination of buffaloes, excreting larvae in their milk, remained negative throughout, while their calves became positive for *N. vitulorum* eggs, from 27–42 days after birth. The passage of eggs in the faeces of young buffalo calves from positive dams confirms the transmammary route of infection for this parasite.

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Hissar, *April* 8, 1975.

O. P. Gautam.
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D. K. Singh.

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1. Olsen, O. W. and Lyons, E. T., *J. Parasit.*, 1965, **51** (5), 689.

2. Stone, W. M. and Girardeau, M. H., *Vet. Med.*, 1967, **62**, 252.
3. Warren, E. G., *Aust. vet. J.*, 1969, **45**, 388.
4. Shoho, C., *Helm. Abst.* (Series A), 1971, **40** (Pt. 2), No. 2971.
5. Srivastava, H. D. and Mehra, K. N., *Proc.* 42*nd Indian Sci. Congr.*, 1955, Pt. III, 354.

Leaf Blight of Cotton Caused by *Macrophomina phaseolina*

During August and September 1975 a hitherto unknown leaf blight and leaf spot of cotton was observed in Haryana Agricultural University Experimental Farm. Blight symptoms were characterised by yellowing or browning of leaves which invariably initiated from apical ends and was followed by impregnation with minute dot like pycnidia. There was no pronounced demarcation between blighted and healthy lamina. Leaf spots were mostly confined to margins of the leaves. They were elongated and confluent with dark coloured border separating them with healthy tissue. The infected lamina were densely studded with fungal pycnidia as in case of leaf blight described above. Both types of symptoms were sometimes observed on the same leaf.

Microscopic examination of the pychidia from blighted and spotted leaves revealed the causative fungus as *Macrophomina phaseolina* (Tassi) Goid. Isolations from infected tissues also consistantly yielded its sclerotial form *Rhizoctonia bataticola* (Taub.) Butler, which is characteristic of this pycnidial fungus. Pycnidial formation in culture has, however, been reported recently by Chidambaran and Mathur[1], under specialisaed condition.

Pathogenicity of the fungus was confirmed ky spraying conidial suspension prepared by homogenising pycnidia from infected leaves as well as by inoculating the leaves with sclerotial agar bit taken from young vigorously growing culture, which after placing on the leaves was moistened with a cotton pad. Leaves inoculated above were enveloped with polythene bags atleast for 48 hours for retention of humidity around such leaves. Symptoms were observed 5 days after inoculation either of the methods. Pycnidia on such leaves, however, appeared 12 days after inoculation. Isolations from such leaves yielded sclerotial form of the inoculated pathogen *i.e.*, *Rhizoctonia bataticola*.

Macrophomina phaseolina is a pathogen of world-wide importance and has been reported chiefly to cause root rot and stem rot of many hosts. In recent years it has been reported to attack foliage of colocassia[2], urid[3], straberry[5], and mung[7], but so far there is no previous report of this fungus affecting cotton leaves and the occurrence of its pycnidia over infected lamina. The fungus is, however, well known to cause root rot of cotton in India and its pycnidia too have been observed on naturally affected cotton stems by Sunderaraman[8] and Kulkarni *et al.*[4]. Leaf blight and leaf spot of cotton incited by *Macrophomina phaseolina* and manifestation of its pycnidia over infected lamina, reported in this communication is, therefore, the first report from India or elsewhere.

Our grateful thanks are due to Dr. D. P. Singh, Director of Research, Haryana Agricultural University, Hissar, for his keen interest and encouragement from time to time.

Haryana Agricultural University, M. P. SRIVASTAVA.
Hissar–125004, B. R. MOR.
October 9, 1975.

1. Chidambaran, P. and Mathur, S. B., *Trasn. Brt. Mycol. Soc.*, 1975, **64**, 175.
2. Grover, R. K. and Parashar, R. D., *F. A. O. Plant Prot. Bull.*, 1968, **16**, 7.
3. Jain, N. K. and Khare, M. N., *Mysore J. agric. Sci.*, 1972, **6**, 150.
4. Kulkarni, Y. S., Thirumalachar, M. J. and Patil, M. K., *Indian Cotton Growers Rev.*, 1958, **12** (4), 247.
5. Lele, V. C. and Pathak, H. C., *Indian Phytopath.*, 1965, **19**, 38.
6. Mathur, S. B., *Phytopath. Z.*, 1967, **58**, 137.
7. Philip, C. T., Kartha, K. K., Joshi, R. K. and Nema, K. G., *JNKVV Res. J.*, 1969, **3** (1), 40.
8. Sunderaraman, S., *Administration Report of the Mycologist for the Year* 1930–31, p. 20, (*cf. Abstr. R.A.M.*, 1932, **11**, 223).

Regeneration of Tail in *Caloosia heterocephala* n.sp. (Nematoda: Hemicycliophoridae)

The tail in *Caloosia heterocephala* n.sp. (Rao and Das, 1975)[1] is filiform with a pointed tip in both males and females. In the males, the caudal alae are crenate and extend upto near the tail tip. The tail measures 195–202 μ in females and 84–110 μ in males. In routine survey of upland rice soils *C. heterocephala* with blunt tails were occasionally encountered. In *Hoplolaimus* spp. (Nematoda: Hoplolaimidae) heteromorphism of tails has been reported earlier[2,3]. Since the frequency of *C. heterocephala* with heteromorphic tails was rare in the fields, it was suspected that the blunt tailed specimens could be due to regeneration of the lost tails.

With a view to investigating the regeneration, 50 females of *C. heterocephala* were colonised in distilled water to which was added 0·01% Streptomycin sulphate. Amputation of tails was conducted with sterilised occular scalpel in a sterilisation chamber. The tail was cut off at a distance of abour 50 μ fron tip. Following the amputation, 32 individuals revived and were active. The tails assumed conoid and blunt shape on the third to fifth day after amputation. Inoculation to rice roots (1) confirmed the ability of these nematodes to invade the root meristem. The ability of *C. heterocephala* to regenerate damaged tail, a new report on the behaviour of this nematode, indicate that dimorphism of tails did not exist in fields.

Central Rice Research Institute,
Cuttack 753 006, *November* 28, 1975
J. SATYANARAYANA PRASAD.

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1. Seshagiri Rao, Y. and Mohanadas, C., *Nematologica*, 1975, **21** (In press).
2. Minton, N. A. and Golden, A. M., *Ibid.*, 1968, **14**, 11.
3. Gupta, P. and Edward, J. C., *Indian J. Nematol.*, 1974, **4**, 100.

A Non-Citrus Host for Citrus Mosaic Virus

A mosaic disease of sathgudi [*Citrus sinensis* (L.) Osb.] from Andhra Pradesh has been described by Reddy *et al.*[1] This note reports successful transmission of citrus mosaic to a non-citrus host *i.e.*, *Hibiscus rosasinensis* (L.).

Six plants of *H. rosasinensis* were graft-inoculated using the inoculum from original citur mosaic by leaf patch grafting. One square centimeter of mosaic infected leaf was introduced into the test plant liftin the bark flap. Immediately after this, the bark flap was sealed with alkathene tape. All the plants exhibited perceptible mosaic symptoms similar to sathgudi mosaic after an incubation of 10 months. Using the inoculum from these, *H. rosasinensis plants* graft-inoculations were done on 4 plants of *H. rosasinensis* and 4 sathgudi plants. Mosaic symptoms were observed in both the best varieties. The control plants remained normal. This is the first report of a non-citrus host for citrus mosaic virus.

Citrus Die-back Project,
S.V. Agricultural College,
Tirupati–517502, A.P., *November*, 4, 1975.
V. DAKSHINAMURTI.
G. SIDDALINGA REDDY.

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1. Reddy, G. S., Moses, G. J., Murti, V. D., Reddy, K. D. and Reddy, V. R. K., Occurrence of mosaic and infectious variegation on sweet orange in Andhra Pradesh, *Abs. 3rd Intern. Symp. Subtropical and Tropical Hort.*, Bangalore, 1972, pp. 130.

Leaf Blight of Cowpea (*Vigna sinensis* Endl.) Caused by *Sclerotium rolfsii* Sacc.

Mortality upto 20% of 5–10 day old seedlings of Co. 2 cowpea, an important pulse crop of India was observed in experimental sowings at the Coimbatore Campus of Tamil Nadu Agricultural University due to seedling blight. The first symptoms appeared in the form of minute brown water-soaked dots which expand into small irregular necrotic spots of 2–3 mm size. With the advance of the disease, the necrotic tissues resulted in the formation of shot holes. In severe cases, the entire leaf was as blighted leading to the death of the plants. In some plants, the cotyledonary leaves showed rotting and faild to open leading to the death of the seedlings.

The pathogen was isolated on oats agar medium both from the leaf spot and the blighted lesions. The fungus formed milky white mycelium with characteristic radial growth and sclerotia were produced within a week. The size of sclerotia ranged from 546 to 1092 μ in diameter and it was identified as *Sclerotium rolfsii* Sacc. When seedlings of Co. 2 cowpea were inoculated with the fungus isolated, typical symptoms developed as stated above. The fungus was reisolated from such inoculated infested leaves. Characteristic symptoms were obtained with both, mycelial bit and sclerotial inoculations. Sclerotial inoculations resulted in typical leaf spots.

In addition to *Vigna sinensis*, the fungus was found to be pathogenic on *Arachis hypogaea* L., *Dolichos lab lab*, *Phaseolus aureus*, *Phaseolus mungo* and *Glycine max*, causing similar symptoms within three days of inoculation. Natural infection of *A. hypogaea* and *D. lab lab* by the pathogen was also observed. The isolates obtained from *V. sinensis*, *A. hypogaea* and *D. lab lab* were cross inoculable indicating that the isolates are indentical.

Sclerotium rolfsii is an ubiquitous fungus causing root rot and wilt of a variety of crops.[1]. *S. rolfsii* has been known to cause root and stem rot of *Vigna sinensis* and *Arachis hypogaea* L.[2], But the induction of leaf spot on cowpea has been reported for the first time. However, this fungus has been known to cause leaf spot of Urid (*Phaseolus mungo*) and *Arachis hypogaea*[3,4],

Deprtment of Plant Pathology,
Tamil Nadu Agricultural University, Coimbatore–3,
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C. L. SUBRAMANIAM.

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1. Ray, A. K., *Sci. and Cult.*, 1973, **39**. 319.
2. Subramaniam, K. S., "Studies on Sclerotial Root Rot Disease of Groundnut (*Arachis hypogaea* L.) Caused by *Sclerotium rolfsii* Sacc.," *M.Sc. (Ag.) Thesis*, Madras University, 1960, p. 59.
3. Mishra, R. P., Khare, M. N. and Chand, J. N., *Indian Phytopath.*, 1971, **24**, 609.
4. — and —, *Ibid.*, 1970, **23**. 706.

REVIEWS AND NOTICES OF BOOKS

Mathematical Programming *via* Augmented Lagrangians. By D. A. Pierre and M. J. Lowe. (Addison-Wesley, Reading, Mass., USA), 1975. Pp. xxi + 436. Price : $ 24.50.

This book is mainly based on the authors' own contribution on multiplier algorithms for nonlinear programming and refers to the latest development in the field. It has seven chapters and several appendices, details of which are given below. The book is self-contained and is very readable with lucid presentation. It can be used as a text book for a special course or lecture series.

Chapter 1: Deals with preliminary concepts and problem statements. Reviews the two general approaches to the constrained maximization namely, (i) the function modification followed by unconstrained optimization and (ii) Direction modification without altering the function (of which very little is available in the existing literature), the former method (i) in detail.

Chapter 2: Devotes to the definitions of differentiability, continuity, Taylor series expansion in *n* variables, and the necessary and sufficient conditions for constrained and unconstrained maxima. Some examples are worked out to illustrate these points.

Chapter 3: A small chapter on sensitivity in which microscopic and macroscopic sensitivity are introduced with examples.

Chapter 4: This is the heart of the book. Three new theorems (author's own theorems) are introduecd. A new Lagrangian called the augmented Langrangian La is defined and the above mentioned theorems prove some interesting properties of this La. An algorithm is developed based on La. A concept called Local Duality is introduced forming a basis for updating the weighting factor in L_a.

Chapter 5: Deals in detail the compur implementation of the algorithm of Chapter 4 under the name LPNLP. The main routines are given incorporating 4 different modes/options for the unconstrained minimization phase. These options are the Davidon Fletcher Powel (DFP) method, the DFP with reset, self scaling (SS) and SS with reset.

Chapter 6: 17 test problems on constrained and unconstrained are worked out using the different options of LPNLP and compared.

Chapter 7: Some practical problems like circuit, and geometric problems are formulated and solved.

About 100 pages are devoted to appendices dealing with general notation, instructions for using LP and NLP, the computer listing of the Fortran program, modified Lagrangian form, matrix operations including programs for matrix operations and summary of unconstrained search methods.

Extensive references are also given.

B. L. DEEKSHATULU.

Techniques and Methods of Polymer Evaluation. Vol. 4: *Polymer Molecular Weights.* Part II. Edited by Philip E. Slade, Jr. (Marcel Dekker, Inc., New York), 1975. Pp. ix + 337 (287–623). Price $ 27.50.

Part II of the book entitled 'Polymer Molecular Weights' consists of three Chapters—Chapters 6, 7 and 8. Chapter 6 deals with 'Gel Permeation Chromatography'. In the past 12–15 years this method has become the most widely used method for the determination of molecular weight and molecular weight distribution of polymers. Considerable progress has been made in developing the method, determining its limitations, and in overcoming the accompanying difficulties that arise. This chapter reviews in detail the previous work and deals at length with the theory, instrumentation and operation, and applications.

'Viscometric Methods of Studying Molecular Weight and Molecular Weight Distribution' is the topic of Chapter 7. Measurement of the viscosity of polymer systems is one of the most popular methods for obtaining information about the molecular weight and molecular weight distribution on account of the relatively inexpensive nature of the apparatus required as compared with other methods. Starting with the definition of viscosity, this chapter discusses in detail the viscosity of both dilute, and concentrated polymer solutions, experimental methods, and the extrapolation methods.

'Sedimentation Techniques' form the subject-matter of Chapter 8. Since the development of the first analytical ultracentrifuge by Svedberg and his coworkers in the twenties, the technique of sedimentation analysis has been perfected to a great extent. Although the ultracentrifuge has become a standard tool for determining the molecular weight in the field of Biochemistry, yet it is a valuable aid for characterising synthetic polymers. This chapter deals with the instrumentation, and the two types of sedimentation analysis, *viz.*, sedimentation velocity, and sedimentation equilibrium analysis.

At the end of each chapter symbols that have been used in the chapter, and references are listed. Each chapter is complete by itself, as it deals with the concerned theory, instrument design, data analysis, etc.

This, two part book, on 'Polymer Molecular Weights' deals at length with all the techniques and the methods that are now widely used for polymer evaluation and hence forms a ready and important source of information on molecular weight measurement for polymer chemists, physicists, and all the scientists handling polymeric materials.

V. KALPAGAM.

Energy: Resources, Demand and Conservation. By Chaman Kaskari (Tata Mcgraw-Hill Publication), 1975. Pp, 231. Price Rs. 36·00).

Energy crisis is a multidimensional global problem involving the whole human race, but it has more seriously affected the economic activity of all developing countries such as India, which have economics dependent on large quantities of imported oil. The hike in oil prices has not only sky rocketed the price of fertilizers, but has led to its non-availability in sufficient quantities in world markets, since production has been affected by high oil prices. This in turn has affected food production creating a grave situation for millions of the world's under nourished poor.

It is, therefore, not surprising that the problem of energy—its resources, demand and conservation is engaging the attention of most of the nations. Considerable amount of literature on energy has been published during the last two or three years, but the book under review is perhaps the first one to have been written from the point of view of a developing country like India.

The book has been divided into three sections : The first—" Introduction, energy resources and demand "—(Chapters 1–5) deal with the energy measuring units, the effect of energy on the economic property of nations, the pattern of energy consumption, Indian and world fossil fuel energy sources.

" Sectorial energy Demand and Conservation" form the second section (Chapters 6–14) in which, the energy problems of India, energy consumption in different sectors of the Indian economy, potential of globar plants, electric power generation including nuclear power and the techniques of energy conservation are discussed.

" Other Energy Resources " (Chapters 15–21) constitute the subject-matter of the third section wherein the possibility of producing energy from waste and scrap, wind, geothermal sources and the sun are described and the techniques of saving energy by the use of total energy systems are suggested.

The book deals in a fairly comprehensive manner of various problems associated with energy in India, and I have no doubt that both technical and non-technical men interested or involved with these problems, will find the book interesting and useful.

The get-up of the book is attractive and the price reasonable. H. N. RAMACHANDRA RAO.

Elements of Economic Entomology. By B. Vasantharaj, David and T. Kumaraswami. (Popular Book Depot, Madras), 1975. Pp. 507. Price Rs. 25 or Rs. 30. Library Edition, $ 10.00 or £ 4.00 (Foreign).

Agricultural education in India is making rapid strides in recent years, and starting of Agricultural Universities in different States has contributed immensely for this spread. Economic entomology constitutes an important component of agricultural education and there are at present hardly any book for the undergraduate student in agriculture for his reference. In this context the present book by David and Kumaraswami is most welcome.

The 26 chapters included in the book are appropriately grouped under five parts. Part I gives a brief account of the history of entomology in India, and also of popular classification of insects.

Part II which consists of over 175 pages is devoted to pests of agricultural, forestry, veterinary and medical importance, of which crop pests occupy over 130 pages. Brief account of bionomics including diagnostic characters of important pests along with control measures and a few diagrams are provided under each crop. In addition, a list of less important or casual insects is given in the end under each crop. Some of the recent recommendations are also included in the suggested control measures, though these include applications of the more persistent insecticides, like DDT and BHC on crops nearing harvest and on vegetables and fruits. Different productive and helpful insects are dealt with in the two chapters which form Part III. Honey bees, silkworms and lac insects are treated in a fairly detailed manner. Interesting account of pollinators, parasites, predators, scavengers, etc., is also given.

Principles and methods of pest control which form the subject-matter of Part IV include among the different conventional methods, the recent tools like the genetic methods, hormones, and pheromones, and finally an integrated approach in pest control. A fairly good review of the properties and action of different groups of insecticides is provided. This chapter also includes information on tolerance limits of some of the pesticides, resistance to insecticides, pollution problems, precautions and antidotes, and a compatibility chart. The chapter on pesticide appliances gives an illustrated account of different types of equipment including aircraft application of pesticides.

Part V is devoted to brief account of different non-insect pests of crops and domestic animals. The two appendices relating to conversion of weights and measures and a ready reckoner for preparation of spray solutions from proprietory insecticides will be useful.

A revision of this book when undertaken may take note of some of the lacunae: History of Indian Entomology may include a history on instruction in Entomology in Indian Universities. The figures, especially of agricultural pests serve very little to illustrate what is desired. A constancy in citing reference to names of insects is highly desirable. A few misspellings here and there may be avoided.

The book serves as a useful reference material to agricultural students and teachers, and the price fixed for a book of this size should be considered as well within the reach of the student community.

G. P. C.

Annual Review of Astronomy and Astraphysics (Vol.13). Edited by G. R. Burbidge, D. Layzer and J. G. Phillips. (Annual Reviews, Inc., 4139 El Camino Way, Palo Alto, California 94306), 1975. Pp. 557. Price: U.S.A. $ 15.00; elsewhere $ 15.50.

The present volume contains fifteen review articles covering a wide range of subjects. On the obsere vational side van de Kamp writes on astronomy whil- Peterson gives a comprehensive account of instrumental techniques in X-ray astronomy. The use of on-line digital computers for telescope control, data acquisition and data reduction has become widespread in observatories round the world in recent years. Robinson's article on the subject describes the various systems in use and will prove usefnl to workers trying to set up new control equipment for their telescopes.

Spectroscopic and photometric studies with new equipment on powerful telescopes have increased the scope of population classification in galaxies and have provided additional information on their composition and evolution. Since most of the work has been confined to the local group, van den Bergh's review on the subject concentrates on the members of the local group of galaxies although some work on extra local group galaxies has also been reported. A related article by Peimbert discusses abundance determination of the gaseous component of galaxies as obtained from the studies of HII regions and planetary nebulae. One of the basic data obtained from these studies is the helium abundance y. Peimberts analysis of the data yields a value for the primeval helium abundance which coupled with the standard big-bang model indicates that we live in an open universe.

The young subject of experimental stellar dynamics has been reviewed for the first time in this series by Aarseth and Lecar. Starting with a general statement of the N-body problem the authors have discussed various astronomical contexts where these simulations find application, *e.g.*, clusters of stars and clusters of galaxies.

Since the beginning of ultraviolet studies about a decade ago, the observations in this field have revealed very interesting things about stars and interstellar gas. Copernicus, launched three years ago has provided a wealth of information regarding abundances and the state of ionization of the intersteallr medium. Spitzer and Jenkins discuss and interpret the data analysed to date and point out significant difference with existing ideas derived mostly from radio observations.

The late stages of stellar evolution are beset with many uncertainties and one of these is a correct evaluation of the effect of neutrinos on the physics of stellar interiors. It appears from Barket's review that theoreticians are now taking the hypothetical electron-neutrino coupling more seriously, and most models of the late phases of stellar evolution include and discuss the role of neutrinos. His review has the necessary skepticism, but still covers all the probable situations where these massless fermions make their presence falt.

Beyond the late phases of stellar evolution is the eventual demise of a star and one of the final configurations, a star may reduce to, is a black hole. Eardley and Press have described the formation of black holes from stellar collapse and the subsequent interaction of the hole with its environment. This article also includes a section on black holes in binary systems and possible identifications, *e.g.*, Cyg X-1. Canuto's sequel on the equation of state at Ultrahigh densities (the first article was published in *Ann. Rev.* Vol. 12), is relevant again to interiors of stars in very advanced stages of evolution and the density range covered in the present article ($P \geqslant 10^{14}$ gm cm^{-3}) is only important in the case of neutron star matter. Canuto has summarised the equation of state in various density ranges in the form of tables which can conveniently be used when calculations on very dense interiors are undertaken.

The problems of star formation and pre-main sequence evolution are as fundamental as those in the later phases of evolution. With the advent of infrared astronomy and the extension of radio observations, significant progress has been made in our understanding of very young objects and their association with the interstellar medium. Detailed theoretical calculations have also been started to trace the early life of stars. The article by Strom *et al.* gives an excellent overview of both the theoretical work and the present state of observations. As they have rightly pointed out our knowledge is particularly lacking in the prestellar physical processes, and empirical insight rather than theoretical speculation should prove rewarding.

Besides the ones described above, this volume includes articles on radio surveys, high velocity neutral hydrogen, the emission mechanisms of pulsars and a sequel by Fowler, Caughlan and Zimmerman to their first article on thermonuclear reaction rates published in this series in 1967.

D. C. V. MALLIK

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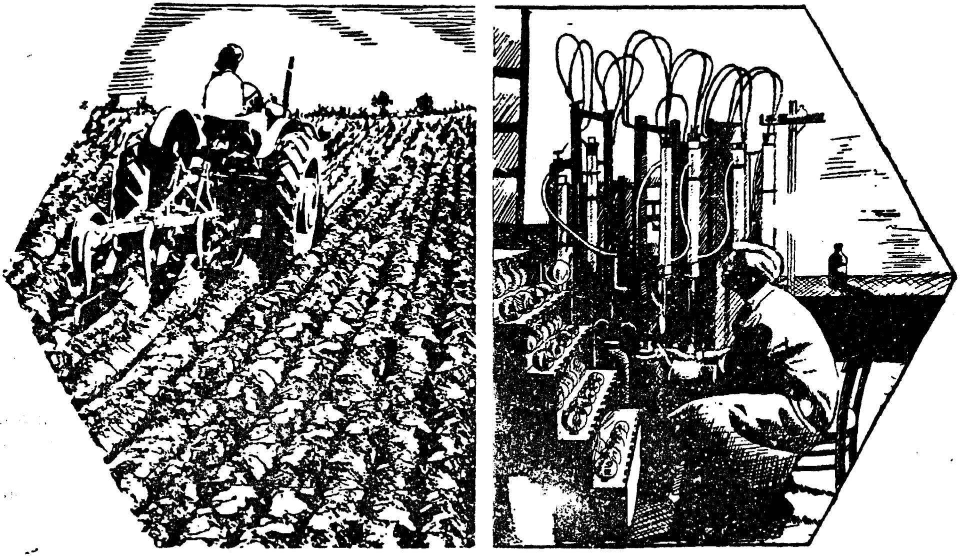

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IS COMPRESSIBILITY AN IONIC PROPERTY?*

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Abstract

A novel approach to the formulation of the repulsion potential of ions in ionic crystals has been attempted. The repulsion potential is postulated to be the sum of contributions from the individual ions, these contributions being proportional to the coordination numbers of the ions. For the specific exponential form of the ionic compression energy [A exp $(-r/\rho)$] the two parameters A and ρ have been evaluated for 5 alkali and 4 halogen ions to fit the behaviour of 20 alkali halides. Not only is the number of parameters (18) used in this theory less than that (26) in comparable existing ones but the fit also is better. The same parameters fit the data of the alkali halides, (*a*) at atmospheric pressure, (*b*) in the pressure range 0–45 kb wherever data is available and also (*c*) for both the low pressure NaCl and the high pressure CsCl forms when such transformations occur. The important feature in this approach is that the parameters evaluated for an ion in one compound in one crystal structure can be used directly for the same ion in any other crystal of any structure at any pressure.

Introduction

THE concept of an ion as an incompressible sphere with a specific radius has proved very useful in many fields, particularly so in the crystallography of inorganic substances[1–5]. One is therefore tempted to enquire whether more complex properties like compressibility could also be ion-dependent.

Any crystal is stable because of the balance between attractive and repulsive forces in it. The attractive forces in an ionic crystal mainly consist of electrostatic coulomb forces and dispersion forces arising from van der Waals' dipole-dipole and dipole-quadrupole interactions. The attractive interactions can be calculated fairly accurately. They involve lattice sums for which powerful methods have been evolved (*e.g.*, Ewald[6]). The repulsion forces between ions are known to arise from the overlap of electronic clouds of the neighbouring ions. However, the actual evaluation of these from first principles is extremely difficult and has been rarely attempted[7]. It is therefore customary to use empirical functions to represent the repulsion interaction (*see* Tosi[8] for a review). We can write the lattice energy per molecule of say a binary ionic crystal in terms of the nearest neighbour distance r as

$$W_L(r) = -\frac{\alpha e^2}{r} - \frac{C}{r^6} - \frac{D}{r^8} + W_{\text{rep}}(r) \qquad (1)$$

where the first three terms constitute the attractive energy and describe respectively the Madelung electrostatic, the dipole-dipole and the dipole-quadrupole interaction energies, and $W_{\text{rep}}(r)$ is the repulsion energy.

The idea of an ion as a separate entity in an ionic crystal cannot be rigorously justified on the basis of quantum mechanics. But accurate electron density maps of ionic crystals[9–11] clearly show that the electrons of each ion are well separated with regions of near zero electron density between the ions giving a physical substance to the concept of the ionic radius. However, considering the ions as hard incompressible spheres, with fixed radii, is too restrictive. We present here an attempt to widen our concept of the ion and treat it as a compressible sphere.

Repulsion—A Result of Ionic Compression

The fundamental postulate we make is that the repulsion energy $W_{\text{rep}}(r)$ in eqn. (1) can be written as the sum of the contributions from the two ions in the form

$$W_{\text{rep}}(r) = W_+(r_+) + W_-(r_-) \qquad (2)$$

where r_+ and r_- are the radii of the two ions. The implications of eqn. (2) are:

(*a*) The ion has an internal energy [*viz.*, $W_+(r_+)$ or $W_-(r_-)$] which is a function of its size, the ion having less energy at large volumes. Figure 1 (full line) shows the type of function that is envisaged for this energy.

(*b*) When the ion is compressed by a pressure, a reaction is set up and the ion reaches an equilibrium size (r_+° in Fig. 1) at which its total energy is minimum.

(*c*) The internal energy of the ion does not depend on the agency that compresses it. For instance, the neighbouring ions of the crystal could be causing the compression. The internal energy of an ion is hence described by the same function in the different crystals in which it occurs [subject to some modifications (discussed later) when the structure type changes].

(*d*) We see from Fig. 1 that a compressed ion reaches an equilibrium size because it resists compression. The basic postulate used in writing down eqn. (2) is that the repulsion between ions arises *solely* from their reaction to compression.

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* Based on an invited talk delivered (by S. R.) at the National Conference in Crystallography, New Delhi, December 4–6, 1975

If the repulsion potential (2) is substituted in the expression (1), the lattice energy of a binary ionic crystal is seen to be a function of three variables, *viz.*, r, r_+ and r_-. However, there are two further relations :

(*a*) Since nearest neighbours are assumed to be in contact

$$r = r_+ + r_-. \qquad (3)$$

(*b*) For any given r, the two ions adjust their sizes so as to minimise their total compression energy. This requires the first differential coefficient of the energies of the two ions to be equal, *i.e.*,

$$\frac{dW_+(r_+)}{dr_+} = \frac{dW_-(r_-)}{dr_-}$$

or

$$W_+'(r_+) = W_-'(r_-). \qquad (4)$$

We can physically picture the neighbouring ions as pushing against each other and so adjusting their radii that the forces they exert on each other are balanced. Equation (4) describes the balancing of the two forces.

Because of eqns (3) and (4), r_+ and r_- are ultimately functions of r and it can be shown that

$$W_+'(r_+) = W_-'(r_-) = \frac{dW_L(r)}{dr} - \frac{\alpha e^2}{r^2} - \frac{6C}{r^7} - \frac{8D}{r^9} = f(r) \text{ (say)} \qquad (5)$$

$f(r)$ has the units of force and is a measure of the compressing pressure acting on the individual ions in the crystal.

$$\frac{dW_L(r)}{dr}$$

is a quantity that can be computed from experimental data (it is proportional to the external pressure applied on the crystal). The next three terms can also be calculated precisely if the constants α, C, D are known, so that $f(r)$ can be calculated for any crystal at any pressure provided the PV data are available.

EXPERIMENTAL JUSTIFICATION OF THE FIRST POSTULATE

In the present picture the ionic radii r_+ and r_- are not *constants* but *variables*. At any given pressure, the ions adjust their radii so as to minimise the total energy given by $W_L(r) + PV$. Even when the externally applied pressure P on the crystal is zero, there is still a pressure acting on the ions [described by $f(r)$] because of the attractive forces among the ions. This attractive force which can be calculated, since all attractive interactions are well understood, varies from one crystal to the other. Even when $P = 0$, the same ion will not have the same radius in two different crystals. This is the basic difference of this theory from the simple hard sphere model. In the present theory, the same ion in two different crystals will have the same radius only when the *internal pressure acting on it* in the two cases are equal. Although the two internal pressures will not normally be equal, it must be possible to select two external pressures P_1 and P_2 such that P_1 acting on the first crystal and P_2 acting on the second crystal both lead to the same internal pressure on the ion. In such a case, the radius of the ion in the two crystals must be equal. This, if verified, will be a strong justification of the postulate that has been made. However, the ionic radius is not an easily accessible quantity even under ordinary conditions, and to obtain it under different pressures would practically be impossible. Fortunately an equally powerful test is possible. Consider four ions A^+, B^+, C^-, D^- and the four crystals they form

$$\underset{\text{I}}{A^+C^-} \qquad \underset{\text{II}}{A^+D^-} \qquad \underset{\text{III}}{B^+C^-} \qquad \underset{\text{IV}}{B^+D^-} \qquad (6)$$

Now four pressures P_{I}, P_{II}, P_{III}, P_{IV} can be calculated such that P_{I} acting on the crystal I, P_{II} acting on the crystal II, etc., lead to the same internal pressure [*i.e.*, the same value of $f(r)$]. Under these conditions, the ion A^+ has the same radius in crystals I and II, the ion C^- has the same radius in crystals I and III, etc. It is then easy to derive from (3) the relationship between the interionic distances given by

$$(r_{\text{I}} + r_{\text{IV}}) - (r_{\text{II}} + r_{\text{III}}) = 0. \qquad (7)$$

Equation (7) can be readily tested as we need only the nearest neighbour distances as a function of pressure in the four crystals. In the case of the alkali halides family of crystals the test shows that the identity (7) is very well satisfied to less than ½% tolerance in fourteen sets of crystals[12].

It is possible to derive another identity similar to (7) connecting compressibilities. The compressibility of an ion depends on the second derivative of its internal energy and is hence a unique function of its radius. This, however, cannot be directly tested since it is not possible to measure the compressibility of an individual ion. What can be measured is the bulk compressibility of the lattice which has contributions from the individual ions *plus* an " attraction contribution " which arises as follows. As the ions get compressed, the interionic distances change, altering, in their turn, the attractive forces. Now define the following function of r for the crystal

$$g(r) = \frac{1}{\dfrac{d^2 W_L(r)}{dr^2} - \dfrac{d^2 W_a(r)}{dr^2}} \qquad (8)$$

where

$$\frac{d^2 W_L(r)}{dr^2}$$

is inversely proportional to the compressibility of the crystal and W_a $(r,$ represents the total of the attractive interactions [the first three terms in the R.H.S. of (1)] in the crystal. It can then be shown that for four crystals I, II, III, IV at pressures P_I, P_{II}, P_{III}, P_{IV} as described in (6), the following identity results:

$$[g(r)_{I} + g(r)_{IV}] - [g(r)_{II} + g(r)_{III}] = 0. \tag{9}$$

This identity also has been tested on the alkali halides and it checks to about 3%.

The two identities (7) and (9) were derived to be a consequence of the postulate that repulsion arises entirely from the existence of a compression energy for individual ions. Their strong verification may hence be taken to be a fairly good justification of the postulate.

THE EFFECT OF COORDINATION ON THE REPULSION ENERGY

One of the essential features of the present approach is that the functions $W_{\pm}(r_{\pm})$ should be capable of being transferred from one crystal to the other. However, this cannot be done directly when the *structure* changes. If we are to introduce compressibility as a property of the ion irrespective of the structure in which it is situated, we must associate with each ion a unique function which is independent of the symmetry of its environment. We achieve this with a second postulate which states that the compression energy of an ion is directly proportional to the number n of nearest neighbours, *i.e.*,

$$W_{\pm}(r_{\pm}) = nh_{\pm}(r_{\pm}) \tag{10}$$

where $h_{\pm}(r_{\pm})$ is a unique function for a given ion, independent of the crystal structure in which it is situated. The relation (10) can be rationalised on the picture that the increase in internal energy arises from the local compression and distortion at the points where an ion "touches" its neighbours. In this picture an ion may be compared to a balloon, with an impenetrable surface skin and a compressible interior. The second postulate implies the following:

(*a*) Suppose a compound exists in two structures having n_1 and n_2 nearest neighbours. Let the nearest neighbour distance r be the same for structure 1 at pressure P_1 and structure 2 at pressure P_2. Then it is easily shown that

$$\frac{n_1}{n_2} = \frac{f_1(r)}{f_2(r)} = \frac{g_2(r)}{g_1(r)} \tag{11}$$

These identities have been tested on the six compounds KCl, KBr, KI, RbCl, RbBr, RbI which transform from the NaCl to the CsCl structure at high pressures[12]. Here again the results are very encouraging indicating that there is a definite physical basis for the second postulate.

(*b*) We shall show shortly that on the basis of this postulate it is quite straightforward to calculate the contribution to the repulsion from the second neighbour interaction.

THE REPULSION FUNCTION FOR INDIVIDUAL IONS

The next step is to evaluate the function $h(r)$ for different ions. Here again some empiricism is required. The internal energy of the ion has to be represented by a suitable function containing a small number of parameters which have to be refined to fit the data. In our studies we have assumed the following function which would approximate the actual behaviour schematically represented in Fig. 1.

$$h_{\pm}(r_{\pm}) = A_{\pm}\, e^{-r_{\pm}/\rho_{\pm}} \tag{12}$$

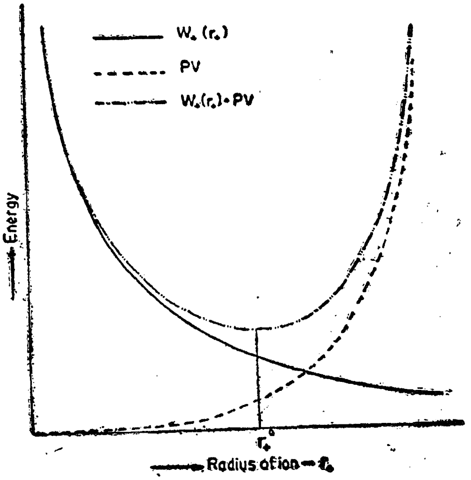

FIG. 1. Schematic graphs of the variation of the internal energy $W_+(r_+)$ of the (positive) ion and the pressure energy PV with r_+ the radius of the ion. The ion takes up a radius r_+° at which the total energy $[W_+(r_+) + PV]$ is a minimum.

where $A_{\pm}$ and $\rho_{\pm}$ are the parameters to be refined, the ± standing for the positive and the negative ion respectively. The failure of this approximation will not in any way invalidate the two postulates made earlier. Another model function [*e.g.*, $h_{\pm}(r_{\pm}) = A_{\pm}/r_{\pm}^{n_{\pm}}$] may possibly give a better description of the ionic compression energy.

Using eqn. (10) we can now write the lattice energy per molecule of a binary ionic crystal as

$$\begin{aligned} W_L(r) = {} & -\frac{\alpha e^2}{r} - \frac{C}{r^6} - \frac{D}{r^8} \\ & + n_1\left[A_+ e^{-r_+/\rho_+} + A_- e^{-r_-/\rho_-}\right] \\ & + n_2\left[A_+ e^{-br/\rho_+} + A_- e^{-br/\rho_-}\right] + PV. \end{aligned} \tag{13}$$

The new term in (11) represents the repulsion energy arising from second neighbours. n_2 is the number of second neighbours and $2br$ is the distance between them. We have used the same compression function for second neighbours also since according to our model, the actual agency compressing an ion is not of consequence. Also we assume that the ionic radius in the direction of the second neighbours is br and so replace $r_\pm$ by br for the second neighbour repulsion terms. This implies that the ion extends to a distance br in the direction of the second neighbours while it extends to r_+ (or r_- as the case may be) in the direction of the first neighbours. In this theory we are compelled to discard the idea of a spherical ion, an aspect strikingly supported by the accurate electron density maps of NaCl obtained by Witte and Wölfel[9] (Fig. 2).

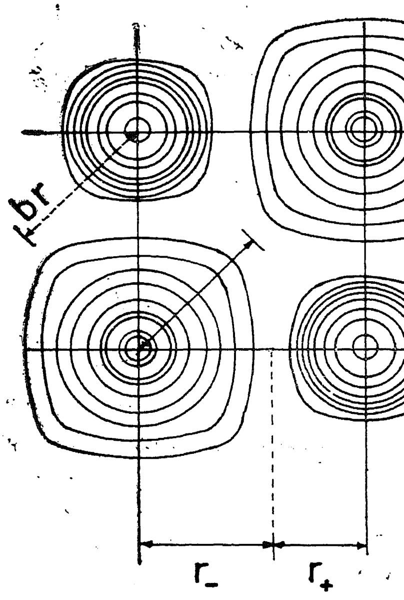

FIG. 2. Electron density plot in the (100) plane of NaCl (after Witte and Wölfel[9]) showing the non-spherical electron distribution in each ion. The ions extend to a distance r_+ and r_- respectively in the direction of the nearest neighbours and to a distance br in the direction of the next nearest neighbours.

The lattice energy function $W_L(r)$ given in (13) was used to fit the repulsion parameters for the five alkali ions Li^+, Na^+, K^+, Rb^+, Cs^+ and the four halogen ions F^-, Cl^-, Br^-, I^-.[13] The available data on the alkali halides were used for this. The lattice energy $W_L(r)$ itself is not very easily measured, neither is it very sensitive to the repulsion function. The equilibrium nearest neighbour distances and the compressibilities which depend on the first and second derivatives of $W_L(r)$ were the data used. These data are available over a range of pressures and the repulsion parameters were refined to fit them over the whole available range. In some cases where there is a phase transformation from NaCl to CsCl structure at high pressures, the data for these new structures were also fitted. This was possible because the second postulate clearly identifies the changes to be made on the repulsion function with change in coordination. Table I gives the final refined parameters for the nine alkali and halogen ions. Table II summarizes the range of validity of these parameters in the alkali halides. The refined parameters fit the experimental data very well justifying the model function used in (10).

TABLE I

Final refined repulsion parameters for the alkali and halogen ions

Ion	A (10^{-9} ergs)	ρ (10^{-9} cm)
Li^+	$7·5 \times 10^{-3}$	1·90
Na^+	$6·4 \times 10^{-2}$	1·815
K^+	0·34	1·848
Rb^+	2·7	1·574
Cs^+	6·5	1·645
F^-	0·60	1·545
Cl^-	3·0	1·67
Br^-	3·0	1·81
I^-	3·0	2·00

TABLE II

The repulsion parameters listed in Table I fit all the available data for the crystals listed below *

LiF	LiCl	LiBr	LiI
NaF	NaCl	NaBr	NaI
KF	KCl	KBr	KI
RbF	RbCl	RbBr	RbI
CsF	*CsCl*	*CsBr*	*CsI*

* Normal type—only atmospheric pressure data available—NaCl type structure.

Normal type underlined—high pressure data available—NaCl type structure.

Normal type underlined and boxed—high pressure data available on both lower and higher sides of a NaCl to CsCl type transformation.

Italics underlined—high pressure data available—CsCl type structure.

CONCLUSION

The picture of repulsion that we have presented is a simplification of a very complex phenomenon but it is our belief that even so simplistic a model is a first step to get an insight into the phenomena underlying the compressibility of ionic crystals. Even if we disregard the physical basis of our arguments and consider the ionic repulsion function we have proposed as one more of the many empirical functions, it has a number of advantages over the existing ones. These are enumerated below.

(1) The latest and most extensive calculation of the repulsion function in alkali halides is the work of Tosi and Fumi[14]. They have employed 26 parameters in their repulsion functions to describe 17 alkali halides (they did not fit CsCl, CsBr, CsI). We require only 18 parameters (one A and one ρ per ion) to fit all the 20 alkali halides listed in Table II.

(2) The repulsion functions of Tosi and Fumi fit the equilibrium nearest neighbour distances with a r.m.s. deviation of 1·45% and compressibilities with 15·4% r.m.s. deviations. The corresponding values for our theory, *viz.*, 0·997% and 6·76% make a very favourable comparison.

(3) Tosi and Fumi attempted only the fitting of atmospheric pressure data. Our repulsion parameters fit the alkali halides (except the fluorides for which reliable high pressure data are not available) in the whole pressure range from 0 to 45 kbars. To our knowledge, no other attempt has so far been made to fit the data over a range of pressures.

(4) Tosi and Fumi[15] refined a new set of parameters whenever a crystal underwent a phase transition. We have eliminated this concept of structure dependent parameters by using the same set of parameters in all phases. This is significant because the attractive interactions for the new phase can all be calculated (the new C and D coefficients are related to the corresponding values in the old phase). Thus, since the repulsion energy is also known, the theory can predict the properties of the new phase.

(5) In our formulation, if the repulsion parameters of an ion have been determined in one crystal of one structure, they can be used for that ion in any other compound of any other structure. When a new compound is encountered, if the parameters of the individual ions have already been refined from other compounds, the repulsion potential for the new compound is completely defined. This potential would, moreover, be valid over a range of pressures. Hence, in principle, all the properties can be calculated. This lends predictive power to our approach. In contrast, almost all earlier attempts fit some crystal-dependent parameters. They can hence never be used to predict the properties of a new system.

(6) As an offshoot of our calculations, we get the "radii" of the ions in the alkali halides. These compare fairly well with the experimental values[13]. Moreover, we also get the variation in the radius of an ion frcm one crystal to the other and with pressure in the same crystal.

We envisage in the future a number of extensions of the above type of calculations to systems other than the alkali halides. We are in particular considering alkaline earth halides and alkali and alkaline earth oxides and chalcogenides. In this context it would be very interesting to test the claim about the predictive power of this approach [point (5) above]. For instance, the repulsion parameters could be calculated for Ba^{++} from the compressibility data on BaF$_2$ using the known parameters of F$^-$. Similarly the parameters of S^{--} could be obtained from the alkali sulphides. Using these new parameters one could check whether the properties cf BaS are well described. It is also hoped to extend the calculations to non-cubic structures if the computational problems are not too formidable and also to ionic radicals. Finally, there is the problem of *explaining* phase transitions. For this, more sophisticated models may be necessary.

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1. Wasastjernas, J., *Soc. Sci. Fenn., Comment. Phys. Math.*, 1923, **1** (37), 38.
2. Fajans, K. and Joos, G., *Z. Physik*, 1923, **23**, 1.
3. Goldschmidt, V. M., *Skrifter Norske Videnskaps—Akad. Oslo, Math Nat.*, 1926, Kl. No. 2.
4. Pauling, L., *Proc. Roy. Soc., London*, 1927, **A114**, 181.
5. Zachariasen, W. H., repored by C Kittel, In *Introduction to Solid State Physics*, Wiley, New York, 1956, Table 3·5.
6. Ewald, P. P., *Ann. Physik* (4), 1921, **64**, 253; *Göttinger Nachr. Math. Phys.*, 1937, Kl. II, **3**, 55.
7. Lõwdin, P. O., *Phil. Mag. Suppl.*, 1956, **5**, 1.
8. Tosi, M. P., In *Solid State Physics* (Edited by F. Seitz and D. Turnbull), Academic Press, New York, 1964, **16**, 1.
9. Witte, H. and Wölfel, E., *Z. Physik Chem.* (Frankfurt) (NS), 1955, **3**, 296.
10. Schoknecht, G., *Z. Naturforsch.*, 1957, **12 a**, 983.
11. Krug, J., Witte, H. and Wölfel, E., *Z. Physik Chem.* (Frankfurt) (NS), 1955, **4**, 36.
12. Narayan, R. and Ramaseshan, S., *Pramāṇa*, 1974, **3**, 143.
13. — and —, *J. Phys. Chem. Solids*, 1976, **37**, 395,
14. Tosi, M. P. and Fumi, F. G., *Ibid.*, 1964, **25**, 45.
15. — and —, *Ibid.*, 1962, **23**, 359.

FILTRATION OF GEOPHYSICAL FIELDS USING EIGEN VECTORS

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ABSTRACT

A digital filter based or the eigen vectors of the correlation matrix of the geophysical field is presented. The advantage of the method is indicated with the help of two field examples.

INTRODUCTION

THE processing of geophysical data involves two basic problems, namely, detection of anomalies and their integrated interpretation. Digital filters are increasingly designed to meet this requirement. Energy filter[1], based on the criterion of the maximisation of the ratio of the energy of the anomaly and the energy of the interferences, enables in particular to resolve problems of detection of oil and gas layers on the basis of the anomalies of the structures in gravity and magnetic prospecting. In the present study anomaly detection procedure is presented by decomposing the geophysical field using orthogonal system of eigen vectors of the correlation matrix.

EIGEN VECTORS CORRELATION MATRIX

Suppose the set of functions $h_i(X_k)$ $i = 1, 2, \cdots m$; $k = 1, 2, \cdots, n$ from an orthogonal system:

(*i.e.*)

$$\sum_{k=1}^{n} h_i(X_k)\, h_j(X_k) = \delta_{ij} \tag{1}$$

where δ_{ij} is the Kronecker's δ and X_k is the station of observation.

Any function of the geophysical field $f(X_k)$ can be expressed as a linear combination of this system of weight functions[2]:

$$f(X_k) = \sum_{i=1}^{m} C_i\, h_i(X_k) \tag{2}$$

where the coefficients C_i are determined by using eq. (1)

$$C_i = \sum_{k=1}^{n} f(X_k)\, h_i(X_k). \tag{3}$$

Weight functions h_i can be found from the minimisation of the mean-square error, given by

$$E_m^2 = \sum_{k=1}^{n} [f(X_k) - \sum_{i=1}^{m} C_i\, h_i(X_k)]^2. \tag{4}$$

Minimisation of the expression on the right hand side of eq. (4) leads to the problem of finding the solution of the system of the following equations:

$$\sum_{k=1}^{n} R_{l-k}\, h_i(X_k) = \lambda_l\, h_l(X_l) \tag{5}$$

$$l = 1, 2, \cdots, n;\quad i = 1, 2, \cdots, m$$

where $[R_{l-k}]$ is the correlation matrix of the initial values of the field. For stationary random function $f(X_k)$ correlation matrix depends only on the shift $(l - k)$. Hence the correlation matrix here can be determined by the autocorrelation functions of the initial values of the geophysical field. However care should be taken to exclude the regional component before taking the initial data for consideration.

Noting that the autocorrelation function is even, we can write the correlation matrix as:

$$[R_{l-k}] = \begin{bmatrix} R(0) & R(1) \cdots R(p-1) \\ R(1) & R(0) \cdots R(p-2) \\ \vdots & \\ R(p-1) & R(p-2) \cdots R(0) \end{bmatrix} \tag{6}$$

where $R(p)$ is given by $R(p) = R'(p)/R'(0)$

$$R'(p) = \frac{1}{n-p} \sum_{i=1}^{n-p} f(X_i)\, f(X_{i-p})$$

and

$$R_{l-k} = R(|l-k|).$$

It is evident that, from eq. (5), the λ_i are eigen values of the matrix (R_{l-k}) and h_i are the corresponding eigen vectors.

ENERGY FILTER

Application of the above theoretical considerations for the detection of the anomaly, on the background of noises, involves only the calculation of the weight functions of the filter based on the criterion of the maximisation of the ratio of the energy of the signal to the power of the interferences[1,3]. Apart from the enhancement of the ratio of signal to noise this criterion takes into account all the useful content of the singal.

The output of any filter is expressed as:

$$Y_k = \sum_{i=1}^{m} h_i\, f(X_{k-i}) \tag{7}$$

where h_i denote the weight function.

Let us suppose $f(X_k) = a_k + n_k$ where $a_k = a(X_k)$ is het anomaly and $n_k = n(X_k)$ the noise.

The energy of the anomaly of the output is defined by:

$$E = \sum_{k=1}^{n+m} C_k^2 \tag{8}$$

where

$$C_k = \sum_{i=1}^{m} h_i\, a_{k-i}. \tag{9}$$

The power of the interferences of the output of the filter is given by

$$P = \sum_{k=1}^{n+m} V_k^2 \tag{10}$$

where

$$V_k = \sum_{i=1}^{m} h_i\, n_{k-i}. \tag{11}$$

The ratio of the energy of the anomaly to the power of interferences is:

$$\lambda = \frac{\dot{E}}{P}. \tag{12}$$

Substituting from eqs. (8), (9), (10), (11) in and eq. (12) simplifying we obtain,

$$\lambda = \frac{\sum_{i=1}^{m} \sum_{j=1}^{m} h_i\, R_{i-j}\, h_j}{\sum_{i=1}^{m} \sum_{j=1}^{m} h_i\, Q_{i-j}\, h_j} \tag{13}$$

where $[R_{i-j}]$ and $[Q_{i-j}]$ are the correlated matrices of the anomaly and interferences respectively.

For maximising λ we differentiate the right hand side of (13) with respect to h_i and equate the derivatives to zero[4]. And thus obtain a system of linear equations as:

$$\sum_{j=1}^{m} [R_{i-j} - \lambda\, Q_{i-j}]\, h_j = 0 \tag{14}$$

$$i = 1, 2, \cdots, m$$

System (14) constitutes the 'generalised eigen value problem'[5] where the signal to noise ratio λ is the eigen value and the filter $[h_i]$ is the associated eigen vector.

If the interferences are uncorrelated and their dispersion is equal to unity,

(*i.e.*)

$$Q(m) = 0, \quad m \neq 0; \quad Q(0) = 1$$

$[Q_{i-j}]$ becomes unit matrix I.

as a result system (14) reduces to:

$$\sum_{j=1}^{m} [R_{i-j} - \lambda I]\, h_j = 0 \tag{15}$$

$$i = 1, 2, \cdots, m$$

Since we want the filter corresponding to maximum value of the signal to noise ratio, we select $[h_i]$ corresponding to $\lambda_{\max}$. Thus from eq. (15),

$$\sum_{j=1}^{m} R_{i-j}\, h_j = \lambda_{\max}\, h_i \tag{16}$$

$$i = 1, 2, \cdots, m$$

System (16) is analogous to system (5). However in case of (16) it is necessary only to find one eigen value of the correlation matrix. Since noise is assumed to be uncorrelated this $\lambda_{\max}$ is calculated from the correlation matrix of the field data, by using iterative techniques which require not much computer time. We then construct the vector $[h_i]$ using eq. (16) and filter the geophysical field using eq. (7). Since the correlation matrix is of controlling importance the dimension of the autocorrelation function is usually limited by the first minimum.

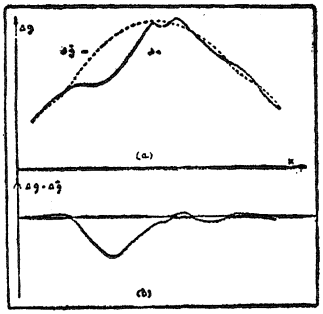

FIG. 1. Filtration of gravity field by latent vectors.

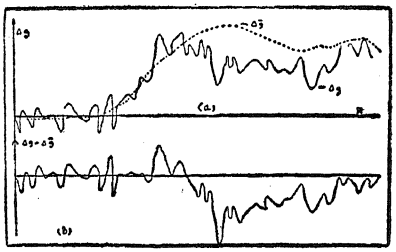

FIG. 2. Filtration of gravity field by latent vectors.

Figures 1 and 2 illustrate the advantage of the processing of geophysical data based on the above method. The initial gravity data ($\triangle g$) from the field surveys for the detection of oil and gas deposits is shown in Figs. 1 (*a*) and 2 (*a*) by a thick line. The filtered gravity field ($\triangle \bar{g}$) is shown by a broken line. The difference curves of the two fields ($\triangle g - \triangle \bar{g}$) are shown in Figs. 1 (*b*) and 2 (*b*) respectively. The anomaly is more pronounced, evidently, in Figs. 1 (*b*) and 2 (*b*).

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1. Trafimova, T. A., Nikitin, A. A., Tarkhov, A. G., "Energetic filter for detection of geophyical anomalies (Russian)," *J. Applied Geophysics*, Moscow, 1974.
2. Churchil, R. V., *Fourier Series and Boundary Value Problems*, McGraw-Hill, New York, 1963, p. 50.
3. Gorbunov, P. N., Beksharipov, K. B. and Klenchin, H. P., "Application of statistical methods for regionalising geophysical fields (Russian)," *J. ASU Kazahstan Geology*, Alma Ata, 1972.
4. Graybill, F. A., *An Introduction to Linear Statistical Methods*, McGraw-Hill, New York, 1961, p. 11.
5. Hildebrand, F. B., *Methods of Applied Mathematics*, Prentice-Hall, Inc., New York, 1952, p. 74.

PROTEIN BONDED POLYMERS: AN APPROACH TO ANTI-THROMBOGENIC SURFACES

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ABSTRACT

Covalent bonding of plasma proteins to suitably derivatized polymer surfaces appears to be a promising method of preparing anti-thrombogenic materials. Plasma protein human serum albumin has been incorporated in derivatized commercial polymers and their blood compatibility evaluated.

PROGRESS in the design and construction of various prosthetic devices, artificial internal organs and heart-lung bypass systems has been severely hampered by the lack of availability of suitable blood compatible materials. All known surfaces other than the vascular interface give rise to blood coagulation on contact with blood. It has been long recognised that the primary event in the clotting process is the denaturation of blood platelets by a foreign surface releasing certain clotting factors[1]. The interaction between the surface of the material and the platelets in the blood is sought to be minimised by modifying the surface using principally three methods : physical, physico-chemical and chemical.

In the purely physical methods of surface modification surface properties of materials like wettability[2] and zeta potential[3] have been investigated in relation to their clotting properties. Gott[4] has tried a novel approach to improve the clotting properties of colloidal graphite by cross-linking the anticoagulant heparin on the surface through a cationic surface active agent. The modified graphite surface obtained by the physico-chimecal method has come to be known as the graphite-benzalkonium-heparin (GBH) surface. The GBH surface has been found to be clot free for 10 days in blood compatibility tests. Later, this technique crosslinking heparin *in situ* has been extended to polymer surfaces[5].

Gott's[4] success with the incorporation of a plasma component on material surfaces clearly shows that the plasma component, being native to blood, would not cause platelet damage when coated on a foreign surface and thus would prevent the initiation of the clotting process. Physical and physico-chemical methods of protein incorporation on a given surface suffer from the disadvantage of weak bonding between the plasma component layer and the polymer molecules. This drawback could be eliminated by covalently linking the plasma components to suitably derivatised polymer and other surfaces. Covalent bonding offers the unique method of preparing water insoluble derivatives which will not go into solution when assayed for activity. Insolubilised proteins find several important applications in biochemical reactions. In the present investigation, however, an attempt has been made to produce thrombo-resistant materials by the incorporation of plasma protein albumin on polymeric materials. The modified polymeric materials have been tested for their blood-compatibility.

MATERIALS AND METHODS

Human serum albumin (HSA) in lyophilized form has been incorporated on polymeric materials like cellulose, polystyrene 6–6 nylon, Dow–Corning silicone elastomer and a commercially available epoxy polymer. In all, six modified surfaces have been prepared using these materials. Four modified surfaces have been prepared by covalently bonding HSA to cellulose, polystyrene and 6–6 nylon. Two modified surfaces have been prepared by physically mixing HSA with Dow–Corning silicone elastomer and epoxy resin. Each of these polymeric materials has been cast into a sheet for evaluating its blood compatibility.

Cellulose—HSA :

Covalent bonding of HSA to *p*-amino-benzyl cellulose was achieved by first preparing the diazonium salt and then adding this to an ice-cooled 1% solution of the plasma protein in borate buffer according to the method of Campbell *et al.*[6].

Polystyrene—HSA :

Polystyrene was first converted to polyaminostyrene by nitration followed by reduction of the surface. The polyaminostyrene surface was then diazotized and HSA coupling was carried out in borate buffer according to the method of Hornby *et al.*[7].

Nylon—HSA :

The technique of Allison *et al.*[8] has been adopted for coupling HSA to 6, 6-nylon. Nylon surface was first hydrolysed by treating with strong (4·5 M) hydrochloric acid. The hydrolysed surface was then treated with glutaraldehyde and HSA in phosphate buffer.

The surfaces have been prepared by mixing HSA with silicone elastomer and epoxy resin and casting the material into sheets,

TABLE I

Control surface		Time for 100% clot formation min.	Protein bonded surface	Time for 100% clot formation min.
6, 6-Nylon	..	7·8	6, 6-Nylon + HSA	16·0
Polystyrene	..	9·5	Polystyrene + HSA	23·0
Epoxy polymer	..	16·0	Epoxy polymer + HSA	29·5
Silicone elastomer	..	31·0	Silicone elastomer + cellulose HSA (32: 1)	59·0
Silicone elastomer	..	do.	Silicone elastomer + cellulose HSA (24: 1)	61·0
Silicone elastomer	..	do.	Silicone elastomer + HSA polymer	74·0

Folin's test[9] and the method of acid hydrolysis[9] have been used to identify the protein in the cellulose HSA conjugate. In the case of polystyrene and nylon protein conjugates infrared spectra have been obtained before and after chemical bonding. A comparison of amide absorption bands at 1650 cm^{-1} to 1550 cm^{-1} showed qualitatively the presence of bonded protein.

The blood compatibility of the protein bonded surfaces was evaluated by the kinetic method of Imai *et al.*[10] using citrated goat blood plasma. Briefly, this method consisted of recalcifying a known volume of plasma on a given surface to initiate the clotting process and of determining the weight of the clot formed after various intervals of time. At the end of a known time interval the clotting process was stopped by diluting the mass with water. The thrombus formed was washed, fixed with formaldehyde, dried and weighed. For each surface a plot of per cent clot formed *versus* time was prepared. Per cent weight of clot is based on the weight of clot formed at infinite time on the same surface. The time taken for 100% clot formation was read from the plot in each case. These times of clotting are compared with values obtained on control surfaces in Table I.

RESULTS AND DISCUSSION

Protein bonded surfaces have shown definite improvement over the control surfaces in their blood compatibility. The highest clotting time obtained was of the order of one hour and the lowest about 15 minutes. Silicone elastomer with HAS polymer and celulose–HSA conjugate gave better results than the other protein bonded polymers. Silicone elastomer is known to be a good blood compatible material and it gave the highest clotting time among the control surfaces. With the modified surfaces clotting times of the order of days have been expected. The low values obtained could be attributed to the possibility of the surface not being completely saturated with protein. This could not be quantitatively checked in the absence of a reliable method for the estimation of the bonded protein. One possible approach to this end would be radio-labelling of the protein.

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1. MacFarlone, R. G., *Nature* (London), 1964, 202, 498.
2. Neubauer, O. and Lampert, H., *Münch med. wschr.*, 1930, 77, 586.
3. Leininger, R. I., *Biophysical Mechanisms in Vascular Homeostasis and Intravascular Thrombosis*, Ed., Sawer, P. N., Appl. Cent. Crofts, 1965, p. 288.
4. Gott, V. L., Whiffen, J. D. and Dutton, R. C., *Science*, 1963, 142, 1297.
5. Lagergren, H. R. and Eriksson, J. C., *Trans. Amer. Soc. Artif. Int. Organs*, 1971, 17, 10.
6. Campbell, D. H., Luescher, E. and Lerman, L. S., *Proc. Natl. Acad. Sci. (U.S.)*, 1951, 37, 575.
7. Hornby, W. E., Fillipuson, H. and MacDonald, A., *FEBS Letters*, 1970, 9, 8.
8. Allison, J. P., Davidson, L., Kitto, G. G. and Hartmann, A., *B.B.R.C.*, 1972, 47, 66.
9. Lowry, O. H., Rosebrough, N. J., Farr, A. L. and Randall, R. J., *J. Biol. Chem.*, 1951, 193, 265.
10. Imai, Y. and Nose, Y., *J. Biomed. Res.*, 1972, 6, 165.

A FLUORESCENT NUCLEOSIDE FROM MYCOBACTERIAL tRNA

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TRANSFER RNAs are rich in modified nucleosides[1]. Apparently, these components are synthesized as secondary modifications of the major constituent nucleosides at the polynucleotide level[2,3]. Modified bases may be present anywhere on the tRNA molecule but their role in the maintenance of structure and function of tRNA is not established. Several tRNAs contain hypermodified derivatives of isopentenyl adenosine next to the 3′-end of the anticodon[1], and the tRNA^{Phe} from several organisms (Yeast, rat liver, ox liver) contains next to the anticodon a hypermodified purine derivative, referred to as 'Y' base and exhibiting characteristic fluorescent properties[4-10]. In this communication, we report the isolation and partial characterization of a new fluorescent nucleoside from mycobacterial tRNA.

Transfer RNAs from *M. smegmatis* and *M. tuberculosis* $H_{37}R_v$ were isolated by the phenol extraction method[11]. The total tRNA was passed through DEAE-cellulose in order to free it from contaminating ribosomal RNA, DNA, oligonucleotides, polyphosphates and polysaccharides. For chemical hydrolysis, the tRNA was treated with 0·5 N KOH for 18 hr at 37° C. The hydrolysate was adjusted to pH 8·5 and digested with alkaline phosphatase to obtain the nucleosides. For enzymatic hydrolysis, the tRNA preparations were separately treated with pancreatic RNase A (10 units/50 $A_{260\,nm}$ units of tRNA) or RNase T_1 (5 units per 15 A_{260nm} units of tRNA). Thin layer chromatography (TLC) was carried out with microcrystalline cellulose. Fluorescent measurements were carried out on a Perkin–Elmar spectrofluorimeter.

The tRNA preparations isolated from two different species of Mycobacteria, *viz.*, *M. tuberculosis* $H_{37}R_v$ and *M. smegmatis* exhibited a high degree of fluorescence. To ascertain whether the fluorescent compound is indeed a part of the tRNA and not a contaminant associated with it, the tRNA samples were hydrolyzed (chemically or enzymatically) and gel filtration on a column of Sephadex C-25 was carried out. While the fluorescence was associated with total tRNA in the void volume when native tRNA was gel filtered, the fluorescent compound also migrated with nucleotides on hydrolysis and fractionation, strongly suggesting that the fluorescent compound may be a constituent of the tRNA. When the total tRNA from *M. smegmatis* was fractionated on a column of benzoylated DEAE-cellulose[12] the fluorescence was associated with the tRNA fractions eluting with high salt and 20% ethanol. Since, these fractions contained a mixture of several tRNAs, as evidenced by the amino acid acceptance, it is necessary to do further fractionation to assign the fluorescent compound to any particular tRNA.

To characterize the nucleosides, the tRNA digestion products were separated by TLC on cellulose (Fig. 1). The nucleosides were eluted using 0·1 N HCl and the ultraviolet spectra were recorded. Tentative identifications of the nucleosides were carried out based on the ultraviolet spectral characteristics at different pH conditions and cochromatography with available nucleosides as reference markers. In addition to the normal

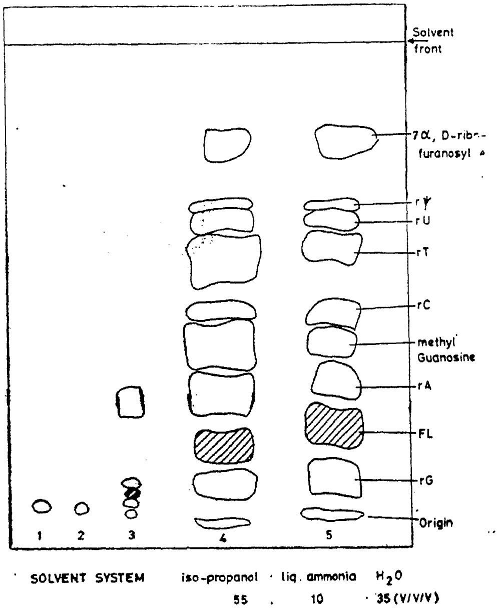

FIG. 1. Thin Layer Chromatography on cellulose. Different tRNA samples, as listed below, were chromatographed using microcrystalline cellulose; 1, *E. coli* total tRNA, untreated; 2, *M. smegmatis* total tRNA, untreated; 3, *M. smegmatis* total tRNA, alkaline hydrolysate; 4, *M. smegmatis* total tRNA, alkali hydrolysis followed by alkaline phosphatase treatment; 5, *M. tuberculosis* $H_{37}R_v$ total tRNA, alkali hydrolysis followed by alkaline phosphatase treatment. (FL = Fluorescent nucleoside.)

constituents of tRNA (U, C, A and G), the modified bases like ribo T, methyl G, pseudo U, and 7, α D-furanosyl A were also present. The fluorescent spot was located near the guanosine residue and showed the characteristic mobility change of nucleotide to nucleoside conversion on treatment with phosphatase. The ultraviolet absorption and the fluorescent spectra of the isolated nucleoside are presented in Figs. 2 and 3. The fluorescent spectra of the isolated compound and the total tRNA were identical.

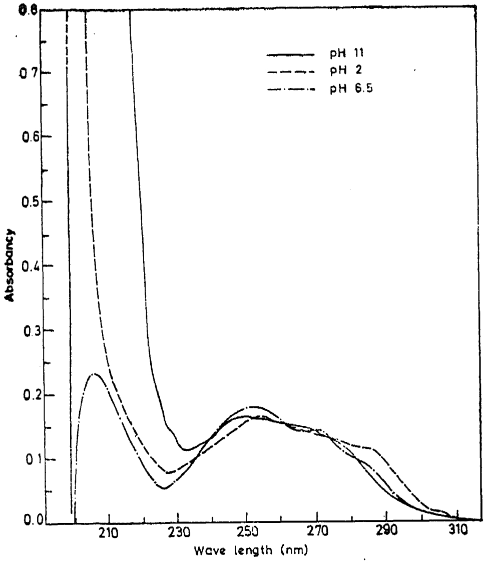

FIG. 2. Ultraviolet absorption spectra of the fluorescent nucleoside from tRNA.

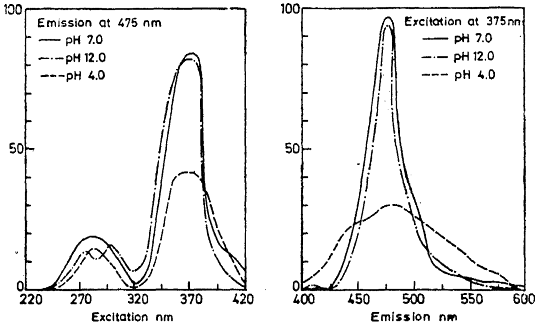

FIG. 3. Fluorescent spectra of nucleoside.

The low mobility of the fluorescent nucleoside on TLC under the conditions used and the patterns of ultraviolet spectra suggested that this might be a modified purine derivative. The nucleoside was however distinctly different from the fluorescent 'Y' base and all other modified nucleosides reported in literature till now. Further experiments for the complete characterization of the nucleoside and its role if any, in the regulation of the tRNA functions, are under way.

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1. Nishimura, S., *Progr. Nucl. Acid. Res. Mol. Biol.*, 1972, 12, 50.
2. Bernhardt, D. and Darnell, J., *J. Mol. Biol.*, 1969, 42, 43.
3. Moshowitz, D. B., *Ibid.*, 1970, **50**, 143.
4. Blobstein, S., Grunberger, D., Weinstein, I. B. and Nakanishi, K., *Biochemistry*, 1973, **12**, 188.
5. Dudock, B. S., Katz, G., Taylor, E. K. and Holley, R. W., *Proc. Natl. Acad. Sci. U.S.A.*, 1969, 62, 941.
6. Fink, L. M., Goto, T. and Weinstein, I. B., *Fed. Proc. Fed. Amer. Soc. Exp. Biol.*, 1969, 28, 409.
7. Nakanishi, K., Furutachi, N., Funamizu, M., Grunberger, D. and Weinstein, I. B., *J. Amer. Chem. Soc.*, 1970, 92, 7617.
8. —, Blobstein, S., Funamizu, M., Furutachi, N., Van Lear, G., Grunberger, D., Lanks, K. W. and Weinstein, I. B., *Nature New Biol.*, 1971, 234, 107.
9. Raj Bhandary, U. L. and Chang, S. H., *J. Biol. Chem.*, 1968, **243**, 598.
10. Thiebe, R. and Zachau, H. G., *Europ. J. Biochem.*, 1968, 5, 546.
11. Capecchi, M. R., *J. Mol. Biol.*, 1966, 21, 173.
12. Deobagkar, D. N., *Ph.D. Thesis*, submitted to the Indian Institute of Science, Bangalore, India, 1975.

LETTERS TO THE EDITOR

SOME CHARACTERISTICS OF 22·8 GeV/c *p* AND 50 GeV/c π^--NUCLEI INTERACTIONS

FROM the number of grey, shower and heavy tracks in an interaction in nuclear emulsion, an attempt has been made to estimate the inelasticity, the number of collisions of the incident particle within the nucleus, the number of secondary particles absorbed inside the nucleus and participating in the further generation of particles at 22·8 GeV/c p and 50 GeV/*c* π^--nuclei interactions.

In this paper a systematic investigation of the interactions of 22·8 GeV/c *p* and 50 GeV/*c* π^--with nuclei of the nuclear emulsion has been made in an attempt to estimate the inelasticity K, the number of collisions ν of the incident particle within the nucleus, the number of created pions absorbed inside the nucleus and the number of created pions participating further in the generation of more secondary particles. For this purpose use has been made of the easily accessible information of the interaction, *viz.*, the number of grey (n_g), heavy (n_h) and shower (n_s) particles only. The results obtained have been compared with the investigations of other workers[1-3] The two show excellent agreement.

The experimental details and the procedure adopted to collect the present sample of interactions has been reported earlier[4-6]. All measurements have been carried out on leitz microscope. To check the biases creeping in the exact determination of the number of particles emitted in an interaction, the procedure adopted earlier has been used. Interactions have been classified into light and heavy ones by following the general practice adopted in nulear emulsions[3,7].

If η be the overall elasticity for all the collisions of the incident nucleon inside the nucleus, an estimate of η is then given by

$$\eta = (E_p - N \,.\, \langle E\rangle)/E_p \qquad (1)$$

where N and $\langle E\rangle$ are the number of particles excluding the primary and the mean energy of the secondary particles created in all collisions. If in an *i*-th collision of the primary particle X_i particles are created with mean energy $\langle E\rangle_i$ the conservation of energy then requires,

$$\eta^{i-1}(1-\eta)\, E_p = X_i \langle E\rangle_i. \qquad (2)$$

Substituting the value of $(1-\eta)$ from (1) we get

$$\eta^{i-1} = X_i \langle E\rangle_i / N \,.\, \langle E\rangle. \qquad (3)$$

Since, the available energy (E_p) for every collision s almost the same[4], equation (3) can be written as,

$$\eta = (1/\nu)^{1/i-1}, \qquad (4)$$

This equation holds good for values of *i* from 1 to ν. The overall value of η can be written as,

$$\bar{\eta} = \{ \sum_{i=1}^{\nu} (1/\nu)^{1/i-1} \}/\nu. \qquad (5)$$

Knowing ν one can determine $\bar{\eta}$ and hence the inelasticity K. In the present experiment the value of ν has been determined as follows:

In the first approximation, the total number N of the secondary particles excluding the persisting primary is given by $1\cdot 5\ (n_s - 1)$. For the case of cascade process ν is estimated from the value of grey (n_g) and heavy (n_h) prongs in the event. It is well known that 75% of the grey tracks are due to recoiling protons. Hence, the total number of recoiling protons and the neutrons becomes $1\cdot 5\, n_g$. The contribution to n_g due to secondary processes inside the nucleus is not ruled out. For some of the pions created in the first collision may either get absorbed or get inelastically scattered creating more pions in their collisions with other nucleons of the nucleus, whereas others may escape without any further interaction. Obviously, these secondary phenomena are statistical in nature and their cross-sections depend among other things, upon the energy of the pions. Therefore, using the experimental information[6] about the pion interactions for a wide energy range, the average behaviour of these processes have been accounted for as follows[9]:

The total number n_{as} of pions which are absorbed or scattered inelastically is given[10] by $n_{as} = n_h/\langle n_h\rangle$, where $\langle n_h\rangle$ is the average number of heavy prongs produced per pion interaction. If a is the fraction of pions which are absorbed $a \times n_{as}$ will be the number of pions which are absorbed, and $(1-a)\, n_{as}$ the number of pions which are inelastically scattered. Using the experimental information given by Chamberlin *et al.*[10], *viz.*, $\langle n_h\rangle = 3$ and $a = 0\cdot 70$. The number of pions absorbed inside the nucleus is given by

$$a\, n_{as} = 0\cdot 70 \times n_h/3 = 0\cdot 23\, n_h$$

and the number of pions making inelastic collisions as,

$$(1\text{–}0\cdot 70)\, n_{as} = (1\text{–}0\cdot 70)\, n_h/3 = 0\cdot 1\, n_h.$$

There should be thus $0\cdot 1\, n_h$ recoiling nucleons due to the inelastic scattering of the created pions. Using this correction the value of ν was taken to be $(1\cdot 5\, n_g - 0\cdot 1\, n_h)$.

One can make an estimate of the number of charged pions n_c, created in a pion nucleon collision by using the logarithmic law[6,11] $n_c = 5\cdot 92\ L_n\ \langle E\rangle - 0\cdot 99$ provided $\langle E\rangle$ the energy of the created pion is known, The number of charged pions created in $0\cdot1\ n_h$ collisions is $(0\cdot1\ n_h) \times [5\cdot92\ \mathrm{L}n\ \langle \mathrm{E} \rangle - 0\cdot99]$. On taking into account the contribution of these secondary processes, the total number of secondary particles N comes out to be,

$$\mathrm{N} = 1\cdot5\,[(n_s - 1) - 0\cdot1\ n_h \times \{5\cdot92\ \mathrm{L}_n\ \langle \mathrm{E} \rangle - 0\cdot99\}] + 0\cdot23\ n_h.$$

The total number of final state particles including the persisting primary is given by $\mathrm{N}' = \mathrm{N} + 1\cdot5$.

The values of the various parameters estimated have been displayed in Table I. Here n_1 is the number of created pions absorbed inside the nucleus, n_2 is the number of pions scattered inelastically inside the nucleus. K, ν and n_{as} have been defined earlier. As expected the number of collisions in case of interactions with heavy nuclei is much greater than that with the light nuclei. This consequently increases the value of K, n_1, n_2 and n_{as}. Further, a comparison of ν at these two energies shows that the cascade mechanism is more prevalent at 22·8 GeV/c than at 50 GeV/c. This clearly supports our earlier result[4], *i.e.*, with increase in the primary energy the tube mechanism takes over the cascade process.

TABLE I

The values of various parameters for light and heavy nuclei

ENERGY GeV	LIGHT n_{as}	LIGHT K	LIGHT n_1	LIGHT n_2	LIGHT ν	HEAVY n_{as}	HEAVY K	HEAVY n_1	HEAVY n_2	HEAVY ν
50 (π – N)	1·51 ± 0·1	0·66 ± 0·06	0·46 ± 0·03	1·05 ± 0·07	1·3 ± 0·06	3·27 ± 0·3	0·73 ± 0·04	0·98 ± 0·08	2·28 ± 0·10	4·69 ± 0·6
22·8 (p — N)	1·38 ± 0·1	0·63 ± 0·05	0·40 ± 0·05	0·98 ± 0·08	1·8 ± 0·15	5·09 ± 0·9	0·73 ± 0·03	1·5 ± 0·09	3·5 ± 0·2	5·63 ± 0·07

The value of K obtained in the present investigation are in good agreement with the ones obtained by K. Rybicke[1] and Gieurla[2] at different energies. Bhowmik and Shivpuri[3] using the experimental information about the energy and momentum of the secondaries have also estimated the value of ν and K in 22·8 GeV/c p-light nuclei interactions. The findings of the present investigation are in good agreement with their[3] observations. From the present investigation and the work carried out earlier[1-2] it seems that at these energies not more than two nucleons participate in the interaction in case of light nuclei and in heavy nuclei not more than six nucleons participate.

The author is grateful to Prof. B. Bhowmik for encouragement and advice and to Dr. S. Singh for useful comments. Thanks are due to Mrs. J. Bhowmik, S. Goel and M. Sen for careful scanning and valuable technical assistance.

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1. Rybicki, K., *Nuovo cim.*, 1967, **49 A**, 233.
2. Gierula, J. L. and Krzydzinski, S., *Nuovo cim.*, 1968, **55 A**, 370.
3. Bhowmik, B and Shivpuri, R. K., *Canad. Jour Phys.*, 1968, **46**, 2557.
4. —, Kaul, S. K. and Singh, S., *First Symposium on High Energy Physics* (Bombay), 1972.
5. —, and Shivpuri, R. K., *Phys. Rev.*, 1967, **160**, 1227.
6. —, Kaul, S. K. and Singh, S., *Nuovo cim. Lett.*, 1973, **8 A**, 503.
7. Artykov, I. Z., Barashenkov, V. S. and Eliseev, S. M., *Nucl. Phys.*, 1968, **B 6**, 11.
8. Perkins, D. H., *Prog. Elem. Part. Cosmic Ray Phys.*, 1960, **5**, 2259.
9. Ekspong, A. G., Frisk, A., Nilsoon, S. and Rome, B. E., *Nucl. Phys.*, 1961, **22**, 353.
10. Chamberlain, O., Goldhaber, G., Jansean, L., Kalogeroponlos, T., Segre, E. and Silberger, R., *Phys. Rev.*, 1959, **113**, 1615.
11. Malhotra, P. K. and Ganguli, S. N., *Phys. Lett.*, 1972, **42 B**, 83.

ROTATIONAL ANALYSIS OF A – X AND B – X SYSTEMS OF INDIUM MONO BROMIDE MOLECULE

THE spectrum of Indium mono bromide molecule in the region λλ 3600–3800 Å was studied in low dispersion by M. Wehrli and E. Miescher (1934) and A. Lakshminarayana and P. B. V. Harnath (1970). Barrett and Mandel (1958) studied the spectrum in absorption in the microwave region and obtained the rotational constants for the ground state. The present work was undertaken to know the exact nature of the excited states involved for

A — X and B — X systems and to find the rotational constants for the excited states also. Rotational analyses of 0, 0 band of B — X system and 0, 0 and 0, 1 bands of A — X system have been carried out.

The spectrum of InBr was excited in a high frequency discharge and was photographed on a two metre plane grating spectrograph at a dispersion of 0·5 Å/mm. The 0, 0 band of B — X system clearly exhibited two heads, inner one of which being a Q head ($\nu_Q = 27381 \cdot 9$ cm^{-1}). The analysis has been carried out by the scheme proposed by Youngner and Winans (1960). The rotational constants obtained from the analysis of 0, 0 band of B — X system are given below :

$InBr^{79}$	$InBr^{81}$
$B_0' = 0 \cdot 05832 (\pm 2)$ cm^{-1}	$B_0' = 0 \cdot 05743 \ (\pm 2)$ cm^{-1}
$D_0' = 2 \cdot 0 (\pm 2) \times 10^{-8}$ cm^{-1}	$D_0' = 2 \cdot 0 (\pm 2) \times 10^{-8}$ cm^{-1}
$B_0'' = 0 \cdot 05560 (\pm 1)$ cm^{-1}	$B_0'' = 0 \cdot 05482 (\pm 2)$ cm^{-1}
$D_0'' = 1 \cdot 6 (\pm 2) \times 10^{-8}$ cm^{-1}	$D_0'' = 1 \cdot 8 \ (\pm 1) \times 10^{-8}$ cm^{-1}

In the A — X system 0, 0 band lines in R branch were resolved upto low J values. The observed rotational isotope shift observed due to Br^{79} asd Br^{81} was in good agreement with calculated values. Analysis has been done using the scheme suggested by Youngner and Winans (1960). The origin of the hand was fixed graphically (26603·16 cm^{-1}). Analysis of 0, 1 band was done by comparing the combination relations for the common upper state. The origins were fixed to be at 26380·19 cm^{-1} and 26378·59 cm^{-1} corresponding to $InBr^{79}$ and $InBr^{81}$.

The rotational constants obtained for A — X system are given below :

$IbBr^{79}$	$InBr^{81}$
$B_0' = 0 \cdot 05835 (\pm 2)$ cm^{-1}	$B_0' = 0 \cdot 05745 (\pm 2)$ cm^{-1}
$D_0' = 1 \cdot 8 (\pm 2) \times 10^{-8}$ cm^{-1}	$D_0' = 2 \cdot 0 (\pm 2) \times 10^{-8}$ cm^{-1}
$B_0'' = 0 \cdot 05562 (\pm 1)$ cm^{-1}	$B_0'' = 0 \cdot 05482 (\pm 2)$ cm^{-1}
$B_1'' = 0 \cdot 05543 (\pm 2)$ cm^{-1}	$B_1'' = 0 \cdot 05463 (\pm 3)$ cm^{-1}
$D_0'' = 1 \cdot 6 (\pm 1) \times 10^{-8}$ cm^{-1}	$D_0'' = 1 \cdot 8 (\pm 2) \times 10^{-8}$ cm^{-1}
$D_1'' = 1 \cdot 8 (\pm 2) \times 10^{-8}$ cm^{-1}	$D_1'' = 1 \cdot 8 (\pm 2) \times 10^{-8}$ cm^{-1}

Values of α_e'' and $\Delta G_{\frac{1}{2}}''$ were determined graphically and are given below along with the microwave data.

	PRESENT STUDY	MICROWAVE DATA [Barret and Mandel (1958)]
$\Delta G_{\frac{1}{2}}$	$221 \cdot 0 (\pm 3)$ cm^{-1}	
α_e	$1 \cdot 90 (\pm 1) \times 10^{-4}$ cm^{-1}	$1 \cdot 903 (\pm 1) \times 10^{-4}$ cm^{-1}
B_e	$0 \cdot 05571 (\pm 1)$ cm^{-1}	$0 \cdot 055710 (\pm 1)$ cm^{-1}
r_e	$2 \cdot 5432 (\pm 2)$ Å	$2 \cdot 5432 (\pm 2)$ Å

The ground state configuration for InBr can be written as $z\sigma^2 y\sigma^2 \omega\pi^4 x\sigma^2$ analogous to similar molecules, giving rise to $^1\Sigma +$ state. Two excited states $^3\Pi$ and $^1\Pi$ arise from the first excited state configuration $z\sigma^2 y\sigma^2 \omega\pi^4 x\sigma v\pi$. The transitions $^3\Pi_0 - X^1\Sigma^+$ and $^3\Pi_1 - X^1\Sigma^+$ are of $^1\Sigma - {}^1\Sigma$ and $^1\Pi - {}^1\Sigma$ type and can thus to be attributed to A — X and B — X systems of InBr respectively.

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---

1. Wehrli, M. and Miescher, E., *Helv. Phys. Acta*, 1934, 7, 298.
2. Barrett, A. H. and Mandel, M., *Phys. Rev.*, 1958, 109, 1572.
3. Youngner, P. and Winans, J. G., *J. Mol. Spectroscopy*, 1960, 4, 23.
4. Lakshminarayana A. and Harnath, P. B. V., *Indian J. Phys.*, 1970, 44, 504.

ULTRASONIC RELAXATION IN 2-BROMO PROPIONIC ACID

Valuable information regarding chemical equilibria can be obtained by ultrasonic relaxation study. Relaxation in carboxylic acids has been studied by Lamb and his co-workers[1,2]. Assuming a monomer-dimer equilibrium they explained the relaxation for acetic and propionic acids. Based on the assumption of monomer-dimer equilibrium, Freedman[3] explained theoretically the results for acetic and propionic acids. But after studying the ultrasonic relaxation in a series of carboxylic acids, Piercy and Rao[4] suggested that the relaxation in them is only due to internal rotation about the carbonyl bond and not due to monomer-dimer reaction.

In order to investigate the effect of halogen substitution in carboxylic acids on relaxation, ultrasonic absorption and velocity have been studied in 2-bromo propionic acid using a standard pulse technique in the frequency range of 3 MHz and temperature range of

30° to 40° C. The temperature of the liquid is maintained constant to within $\pm 0{\cdot}1°$ C by using an ultra-thermostat type NBE. The substance used is of Analar grade and is distilled under vacuum before use. The velocity and absorption measurements are correct to $\pm 0{\cdot}5\%$ and $\pm 8\%$ respectively.

The velocity is found to decrease linearly with increasing temperature as is normal for organic liquids. At each temperature α/f^2 is found to decrease with increasing frequency, where α is the absorption coefficient measured at a frequency f. This indicates a definite relaxation in the frequency region of study

Assuming a single relaxation, the sound absorption (α) can be described by the equation of the form

$$\alpha/f^2 = \mathrm{B} + \mathrm{A}/[1 + (f/f_r{}^2] \qquad (1)$$

where B is the high frequency residual absorption and A is the contribution from the relaxational mechanisms. The excess absorption per wavelength $\alpha'\lambda$ has been computed using the relation

$$\alpha'\lambda = (\alpha/f^2 - \mathrm{B})fc \qquad (2)$$

where c is the velocity of sound. The variation of $\alpha'\lambda$ with frequency is shown in Fig. 1, in which the curves are calculated ones according to equation (1) with

$$\alpha'\lambda = Acf/[1 + (f/f_r)^2] \qquad (3)$$

and the points are experimentally observed values, as computed according to equation (2).

From Fig. 1, it can be seen that the relaxation frequency (f_r) which corresponds to the frequency of the peak excess absorption increases with increasing temperature. The relaxation frequency is centred around 7 MHz at 30° C.

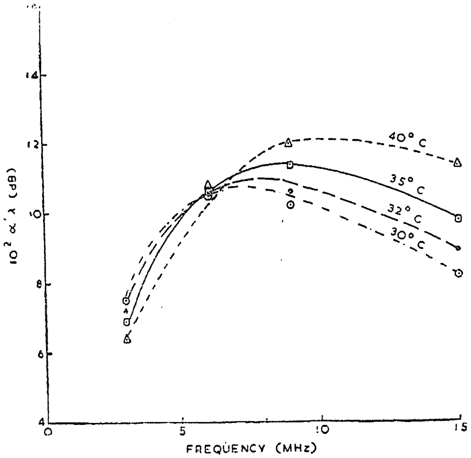

FIG. 1. Frequency dependence of excess absorption per wavelength ($\alpha'\lambda$) for 2-bromo propionic acid at different temperatures.

The variation of the relaxation frequency with concentration is studied in polar solvent acetone and non-polar solvent n-hexane. In both the cases it is found that the relaxation frequency f_r is independent of concentration within the experimental error. This is the characteristic of an intramolecular process and hence the relaxation may be attributed to rotational isomerism about the carbonyl bond as suggested by Piercy and Rao[4].

The authors are grateful to Professor B. Ramachandra Rao for his encouragement and to the Council of Scientific and Industrial Research, New Delhi, for financial assistance.

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1. Lamb, J. and Pinkerton, J. M. M., *Proc. Roy. Soc.*, 1949, **199A**, 114.
2. Huddart, D. H. A. and Lamb, J., *Trans. Faraday Soc.*, 1950, **46**, 540.
3. Freedman, E., *J. Chem. Phys.*, 1953, **21**, 1784.
4. Rao, M. G. S. and Piercy, J. E., *J. Acoust. Soc. Amer.*, 1967, **41**, 1591.

A FURNACE FOR HIGH TEMPERATURE POWDER DIFFRACTOMETRY

HIGH temperature attachment for powder diffractometer is a very useful device for studying several important properties of crystalline materials. A large variety of heating devices have already been described in literature[1]. In this letter we present the design and function of a furnace for use upto 700° C on a Philips powder diffractometer. The salient features of this furnace are : (i) the heater assembly can be easily attached or detached to the sample holder and (ii) the simple cooling device which maintains the diffractometer arm at 30° C when the sample is heated at 700° C.

The cross-sectional view of the furnace fixed to the conventional sample holder used with the Philips diffractometer is shown in Fig. 1. The heating device is fixed below the central opening (a) where the sample under study is kept. The furnace assembly consists of a small porcelian trough (b) 35 × 15 × 8·5 mm in size, the bottom of which is filled with layers of alumina (c) and mica sheets (d). The heating element consists of a 5″ long, 245 WG nichrone wire (e) wound in a ziz-zag fashion and is fitted on the mica sheets (d). The heater leads are taken downward through two vertical side holes (f) made along the edges of the porcelain trough and fused to copper wires connected to the power supply. The filament wire is further covered by a thin layer of mica sheet. The sample is mounted in contact with the mica sheet which provides electrical insulation between the filament wire and the sample. This furnace assembly, as such, could be fixed to the sample holder by means of four corner screws (*g*).

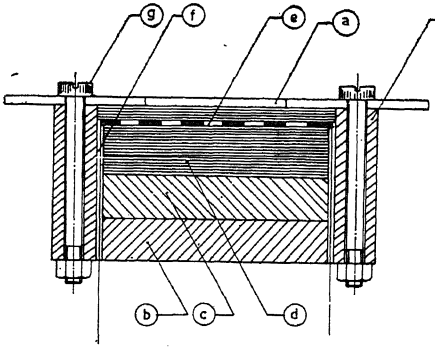

FIG. 1. Cross-sectional view of the furnace fixed to the sample holder.

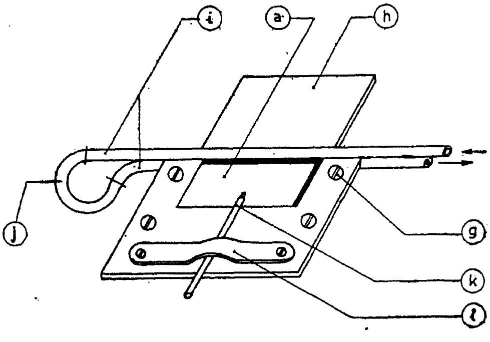

FIG. 2. Cooling and thermocouple arrangement.

The filament wire is heated by stabilised a.c. voltage supplied through a dimmerstat. Fine current control is achieved by using two current-sensitive step-down transformers. With this simple circuit it was possible to control the temperature to an accuracy of ± 5° C. In order to prevent the diffractometer arm (*h*) from getting heated, two 10 cm long copper tubes (*i*) (1·5 mm *i.d.*) are suitably bent and brazed to the lower as well as the upper sides of the brass sample holder. Water which is continuously circulated through the copper tubes linked by a short rubber tubing (*j*) act as the coolant during high temperature experiments. The positioning of the copper tubes is such that they do not hinder the fixing of the sample holder to the diffractometer shaft and they do not also intercept the x-ray beam. With a rate of flow of 350 ml of water per minute, the diffractometer arm could safely be maintained at 30° C when the sample was heated to 700° C. The temperature of the sample is measured by means of a chromel-alumel thermocouple (*k*) which is inserted through a slightly bent metallic strip (*l*) fixed to the top surface of the sample holder. In order to check for thermal gradients along the surface of the sample, the thermocouple could easily be moved under the metallic strip and could be fixed in any position. The output of the thermocouple is fed directly to a digital millivoltmeter which reads the thermal e.m.f. to an accuracy of 0·1 millivolt. At 700° C, the temperature was found to be stable within ± 5° C for a period of about three hours, with a power consumption of about 75 watts. As the sample is fixed directly above the mica-covered filament wire, no appreciable temperature gradient was observed along its surface.

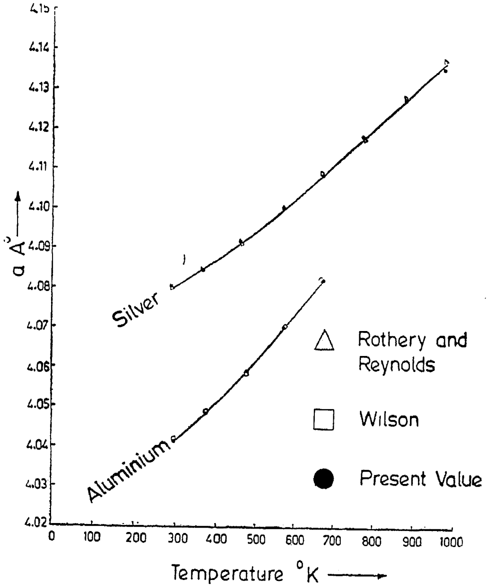

FIG. 3. Temperature variation of lattice parameters of aluminium and silver.

The performance of the heater was tested by measuring the lattice parameters of polycrystalline aluminium upto 400° C and silver upto 700° C. The values plotted in Fig. 3 are found to compare well with those reported in literature[2,3].

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---

1. Mckinstry, H. A., *J. Appl. Phys.*, 1970, **41**, 5074.
2. Wilson, A. J. C., *Proc. Phys. Soc.* (Lond), 1941, **53**, 235.
3. Hume Rothery, W. and Reynolds, P. W., *Proc. Roy. Soc.*, 1938, **167**, 25.

ESTIMATION OF MERCURY IN CHLORITE-CELL LIQUORS

INTRODUCTION

THERE had been increasing use of mercury in the electrolytic production of chlorine, caustic soda and chlorite liquor. The disposal of effluents from these cells poses health-hazards because of the poisonous nature of mercury. Besides being directly piosonous, inorganic mercury let out into lakes and rivers, transforms into organo-mercury compounds in the bodies of fishes, the consumption of which may later cause maladies in human beings. Hence mercury has been an object of attention of the environmental health-laboratories in areas where the mercury/electrolytic-cells operate.

The tolerance limit of mercury in the liquor is 0·01 mg/ml and many cells may be discharging much more than the safety limit. The volumetric and gravimetric methods are rather time-consuming and seldom applicable to micro-gram quantities[1-2]. Spectral analysis by itself or in combination with other techniques, such as electro-deposition[3-7], will involve a large initial capital. Is this context, a rapid and cheap colorimetric estimation as depicted in this paper is welcome.

During the present work, the estimation of micro-gram quantities of mercury in chlorite-cell liquors has been attempted colorimetrically using (*a*) dithizone and (*b*) sulphide methods. The dithizone method was valid between 4–12 p.p.m. of mercury, whereas the sulphide method enabled the estimation between 0–20 p.p.m. making use of working-graphs for the purpose.

EXPERIMENTAL

1. *Preparation of Standards for Working-graph*

The chlorite-cell, liquor, in which the mercury has to be estimated, generally contains a strong solution of the chloride at an acidic pH. To prepare a working-graph, a number of artificial standards are prepared from a 30% sodium chloride solution, keeping the pH at 2 (N/100 with respect to HCl) and mixing aliquots of mercuric chloride solution to give concentrations from 4–32 p.p.m. of mercury.

2. *Mercury Chloride Solution*

AR mercuric chloride, 0·1354 gm was dissolved in 100 ml of 1N HCl. The concentration of this is 1000 p.p.m. of mercury. Aliquots from this solution were added to the sodium chloride solution to prepare a concentration range of 4–32 p.p.m. of mercury.

3. *Dithizone :*

The sample was purified by twice crystallising from chloroform. A 0·01% (weight/volume) solution in chloroform was used in this work.

The following two procedures were adopted for the colorimetric estimation. (*a*) Dithizone extraction and (*b*) Sulphide method.

(*a*) *Dithizone method*.—25 ml of the 'chlorite-cell' liquor prepared as above was extracted twice with 10 ml lots of 0·01% dithizone. The volume was made upto 25 ml. The extracted dithizone complex (orange) containing excess reagent (green) was then measured for its absorption at 500 nm (λ_{max} of mercuric-dithizonate) and at 610 nm (λ_{max} of the reagent). The Beckmann Model B spectrophotometer was used for the measurements. The experiment was conducted as per the procedure described in Sandell[8].

(*b*) *Sulphide method*.—The colorimetric estimation using hydrogen sulphide is preferable for the reasons, that the solubility product of HgS is of the order of 10^{-50} and so the lower limit of estimation can be extended farther down in comparison with the other methods.

25 ml of the 'chlorite-cell' liquor was treated with 0·5 ml of con. HCl and 0·5 gm of mannitol as a protective colliod. Purified H_2S gas was then sent into the cold chlorite-cell liquor for five minutes with agitation. The yellowish-brown colloidal solution obtained is found to be stable for several hours, provided the concentration of mercury did not exceed 20 p.p.m. Beyond this limit, coagulation and precipitation slowly set in and the Beer–Lambert law is not obeyed.

A Dubosque colorimeter was used for colour-matching. A standard solution of concentration 10 p.p.m. was fixed at the scale reading 15 and the readings for the other concentrations at the match-point were read out. These readings were found to follow the Beer–Lambert relationship between limits of 4–20 p.p.m. of mercury. The accuracy of the method is approximately ± 5% at 20 p.p.m. and ± 8% at 4 p.p.m.

TABLE I

Estimation of mercury in chlorite-cell liquors. Sulphide Method

Mercury in p.p.m. Taken	Mercury in p.p.m. Found	% Error
6	6·36	6
8	7·73	3·4
12	12·30	2·5
14	14·71	5
18	17·04	5·3

DISCUSSION

Dithizone Method :

It was found that the percentage transmittance is linear between a concentration of 4–12 p.p.m. of mercury. After 12 p.p.m. the curve flattens. Extraction of mercury by dithizone is incomplete in the presence of high concentrations of sodium chloride in acid medium, as indicated by the formation of a precipitate when H_2S was bubbled through the extract. In the presence of excess chloride ion, mercury forms a complex of the type $(HgCl_4)^{2-}$ with which the dithizone is not able to reach quantitatively. This is also in agreement with a previous report[9]. The instability constants of the mercuric chloride complex and mercuric-dithizonate complex are almost of the same order, being 6×10^{-17} and $3 \cdot 0 \times 10^{-17}$ respectively. Moreover, the mercuric-dithizone complex also suffers photochemical decomposition, despite the addition of a little acetic acid as recommended by the other workers to stabilise the complex-[10]. The colour of the complex is more unstable in the presence of excess of sodium chloride.

It is our observation that the absorption due to the complex is influenced by the excess reagent, dithizone, which has not been mentioned in literature. Under these circumstances, the sulphide method which works better will be welcomed by the industrialists.

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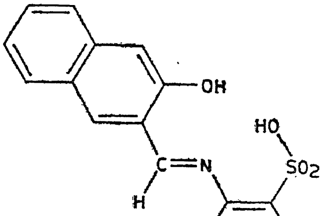
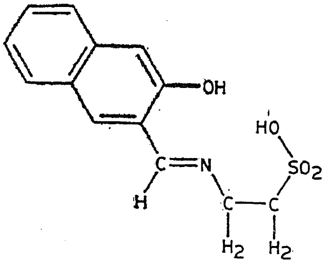
1. Coetzee, J. F., "Mercury," In : Kolthoff, I. M. and Elving, P. J., Eds., *Treatise on Analytical Chemistry,* Part II, Interscience Publishers, John Wiley and Sons, Inc., New York, 1961, 3, 231.
2. Kodama, K., *Methods of Quantitative Inorganic Analysis,* Interscience Publishers, John Wiley and Sons, Inc., New York, 1963, p. 150.
3. *Product Bulletin 400-A,* E.I. du Pont de Nemours and Co., Inc., Wilmington, Del. 19898.
4. Doherty, P. E. and Dorsett, R. S.. *Anal. Chem.,* 1971, 43, 1887.
5. Lee, D. C. and Laufman, C. W., *Ibid.,* 1971, 43, 1127.
6. Schechter, M. M., *J. Ass. Offic. Anal. Chem.,* 1966, 49 (4), 778.
7. Ling, C., *Anal. Chem.,* 1968, 40, 1876.
8. Sandell, E. B., *Colorimetric Determination of Traces of Metals,* Interscience Publishers, Inc., New York, 1944, 3, 322.
9. Irving, H., Andrew, G. and Risdon, E. J., *J. Chem. Soc., London,* Part I, 1949, p. 541.
10. *Encyclopaedia of Industrial Chemical Analysis,* Edited by F. Snell and L. S. Ettre, Interscience, Publishers, John Wiley and Sons, Inc., New York, 1972, 16, 38.

STABILITIES OF SOME BIVALENT METAL-CHELATES OF TRIDENTATE LIGANDS DERIVED FROM 2-HYDROXY-1-NAPHTHALDEHYDE AND β-AMINO ACIDS

THE magnetic and spectral data on N-(2-hydroxy-1-naphthalidene) orthanilic acid (H_2NO) and N-(2-hydroxy-1-napthalidene) taurine (H_2NT) with Co (II), Ni (II), Cu (II), Zn (II), Cd (II) and UO_2 (II) complexes have been reported by us earlier[1]. H_2NO and H_2NT are biprotic tridentates containing sulphonic hydroxyl, phenolic hydroxyl and imino groups (Fig. 1). We have now determined the dissociation constants of these ligands and the stability constants of their metal complexes employing Calvin-Bjerrum pH-titration technique[2,3] in acqueous medium ($\mu = 0 \cdot 1$ M $NaClO_4$) at 30 ± 0·1° C.



N-(2-HYDROXY-1-NAPHTHALIDENE) ORTHANILIC ACID



N-(2-HYDROXY-1-NAPHTHALIDENE) TAURENE

FIG. 1

Precision pH-meter (Type OP–205, No. 837) with glass electrode assembly (accuracy ± ·05 pH) was used for pH-measurements. The ligands were synthesised by the method reported earlier[1]. H_2NO and H_2NT were titrated with 0·1 M sodium hydroxide in the absence and in the presence of metal-ions at 30° C.

TABLE I

Stability constants and free energy changes of H_2NO and H_2NT complexes

Metal		H_2NO Complexes				H_2NT Complexes log K_1
		A*	B	C*	D	
Co (II)	log K_1	5·32	5·27	5·31	5·84	2·82
	log K_2	2·97	2·89	2·97	2·45	..
	log β_2	8·29	8·16	8·28	8·29	..
Ni (II)	log K_1	6·10	5·84	6·09	6·52	2·98
	log K_2	3·45	3·48	3·45	3·03	..
	log β_2	9·55	9·32	9·54	9·55	..
Cu (II)	log K_1	7·55	7·52	7·55	8·27	6·85
	log K_2	4·03	4·29	4·03	3·31	..
	log β_2	11·58	11·81	11·58	11·58	..
UO_2 (II)	log K_1	8·23	8·20	8·22	8·68	7·25
	log K_2	5·62	5·55	5·62	5·17	..
	log β_2	13·85	13·75	13·84	13·85	..
Zn (II)	log K_1	..	..	..	..	2·80
Cd (II)	log K_1	..	..	..	..	2·65

where A = Interpolation at half $\bar{n}$ values; B = Interpolation at various $\bar{n}$ values: C = Schroder method; and D = Correction term method.

* Most suitable methods in this case.

The dissociation constants (pK_1 and pK_2) of H_2NO and H_2NT, obtained by Algebraic and Interpolation at half $\bar{n}_A$ values, were 3·62, 8·07 and 3·37, 9·90, respectively.

The stability constants of the metal-ligand were calculated by using Calvin-Bjerrum pH-titration technique and from the formation curves, $\bar{n}$ *vs* $-\log [A^{-2}]$. The metal chelates of H_2NO with UO_2 (II), Cu (II), Ni (II) and Co (II) were found to form 1 : 2 complexes; Zn (II) and Cd (II) form 1 : 1 complexes only. The formation curves for all the metal –H_2NT systems attain maxima at $\bar{n} < 1\cdot3$ which indicates the formation of 1 : 1 complex. The values of log K_1 and log K_2 in the metal –H_2NO systems were refined by different computational methods[4] and the values obtained are summarized in Table I along with those of the log K_1 values of H_2NT–chelates.

From the perusal of the values it is clear that H_2NO forms more stable complexes than H_2NT. The lower stability of H_2NT complexes may, perhaps, be due to steric factors and the presence of Zwitter-ion. The stability of these complexes follow the order[5]

$$UO_2 (II) > Cu (II) > Ni (II) > Co (II) > Zn (II) > Cd (II).$$

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1. Ozha, D. D., Kaul, K. N. and Mehta, R. K., *Indian J. Chem.* (In press).
2. Calvin, M. and Wilson, K. W., *J. Am. Chem. Soc.*, 1945, 67, 2003.
3. Bjerrum, J., *Metal Ammine Formation in Aqueous Solution* (P. Hasse and Son, Copenhagen). 1941.
4. Irving, H. and Rossotti, H. S., *J. Chem. Soc.*, 1953, p. 3397.
5. — and Williams, R. J. P., *Nature*, London, 1948 162, 746; *J. Chem. Soc.*, 1953, p. 3192.

STUDIES ON BIO-GAS SOURCES

THE production of bio-gas from different excreta (1) Cow, (2) Buffalo, (3) Goat, (4) Sheep, (5) Camel, (6) Horse, (7) Pig, (8) Poultry has been reported in this communication.

The gas production of different waste-materials under room temperature (27–35° C) conditions has been represented in Fig. 1. It may be seen that the

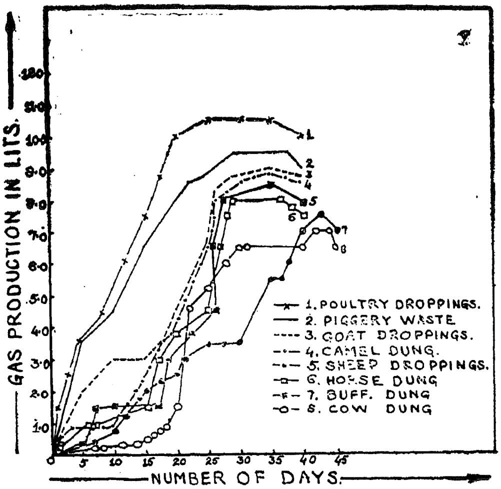

FIG. 1. Gas production behaviour of waste materials.

poultry droppings top the list while cow/buffalo dung remain at the bottom in regard to their gas production. In the case of the poultry droppings gas-production was found very rapid (3 hours) whereas the cow and buffalo dung took as much as one to two weeks time.

The composition of different waste-materials has been given in Table I.

TABLE I
Composition of different waste-materials

Material	Dry Matter %	Volatile Organic Matter (Dry basis) %	Fat %	Crude Protein (Kjeldahl) %	Crude Fibre %	Ash %
1	2	3	4	5	6	7
Cow-dung	18	84	2·4	9·2	21	16
Buffalo-dung	19	83	3·0	9·9	19	17
Horse-dung	33	82	3·0	10·1	25	18
Sheep-droppings	32	81	3·8	17·1	13	19
Camel-dung	32	84	2·2	10·8	16	16
Goat-droppings	32	78	3·2	12·5	13	22
Piggery-waste	45	72	4·5	19·4	26	28
Poultry-droppings	47	65	1·5	26·9	19	34

TABLE II
Production of gas from different waste-materials

Material	Gas production Lits./Kg		
	Raw Material (wet)	Dry Matter	Volatile Organic Matter (dry)
1	2	3	4
Cow-dung	12	60	72
Buffalo-dung	12	65	79
Horse-dung	31	91	111
Sheep-droppings	31	91	113
Camel-dung	34	100	119
Goat-droppings	37	106	135
Piggery-waste	47	107	149
Poultry-droppings	74	126	191

The total gas-production on raw, dry and volatile organic material basis of all the waste-materials as

TABLE III

Composition of gas samples obtained from different waste-materials

SOURCE	No. of trials	COMPOSITION %				
		Co$_2$	O$_2$	CO	CH$_4$	Other Gases
Cow-dung	3	35·8	6·0	0·9	50·1	7·2
Buffalo-dung	3	35·0	6·2	1·0	49·8	8·0
Horse-dung	3	29·6	6·8	1·4	55·2	7·0
Sheep-droppings	3	28·2	6·2	0·8	54·0	10·8
Camel-dung	3	30·0	6·0	0·9	53·0	10·1
Goat-droppings	3	22·8	8·6	0·8	53·2	14·6
Piggery-Waste	3	26·5	7·2	1·8	56·0	8·5
Poultry-droppings	3	25·4	8·5	1·2	56·0	8·9

represented at Table II has been found in accordance with their gas-production behaviour as represented at Fig. 1. Factors like low temperature and scum formation have been found to retard the gas production. Stirring during the experiment was found to accelerate gas production considerably.

The samples of gas obtained from the above waste-materials were analysed employing Orsat Gas Analyser. The comparative composition is represented at Table III.

The gas obtained from the piggery and poultry droppings have been found richer in methane content as compared to the gas obtained from cow/buffalo dung.

The authors express their sincere thanks to the authorities of Haffkine Institute; Zoological Gardens; Bombay Veterinary College; Aarey Milk Colony; Bacon Factory, Borivli; and Regional Poultry Farm, Aarey Milk Colony, for supply of samples. Thanks are also due to the authorities, I.I.T., Powai and Bombay Gas Company, for their laboratory and library facilities.

National Dairy Research Institute,
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A STUDY OF THE EFFECTS OF DDVP (PROPOXURE) ON LABORATORY MOUSE

HARMFUL effects of insecticides on animals including mammals have been studied by several investigators[1-3]. Some insecticides are less potent, and induce overall deleterious effects on biological systems but not mortality. Studies on the effects of these agents on the reproductive system would be of significance to understand the expression of these effects.

DDVP is one of the potent insecticides which does not inflict drastic effects like mutations on mammalian systems. Keeping this in mind, during 1969, for the first time, it was used in the premises of the mice colony of Cancer Research Institute. Though the use of the insecticide was restricted to the area where cages were cleaned and exchanged, it was noticed that a large number of mice in the colony failed to have the expected number of litters. The present study was initiated to ascertain whether DDVP treatment of the mouse cages had contributed to this effect.

Materials and Methods:

Eight to twelve-week old mice of C 57 bl and Swiss/Icrc strains (three females and one male) were placed in each cage with controlled temperature and humidity. When the females were visibly pregnant, they were transferred to DDVP sprayed cages.

The treatment of cages consisted of weekly spraying of DDVP (approximately 2·1–2·4 gm/cage) into each cage. When the females delivered, the treatment was discontinued for two weeks. Spraying of the mothers (F generation) and the weanlings (F_1) was continued after the young were weaned. The treatment was stopped after 10 months in the case of F generation and 8 months in the case of F_1. Fertility of these two groups of mice was assessed on the basis of number of litters delivered by the females.

All animals were maintained until death. Animals which were observed weak or sick were sacrificed, their ovaries or testes dissected for histological study.

Observations :

In the direct treatment group, the first observable effect following three months treatment was ruffled hair, breathing difficulties, diarrhoea and general weakness. The long term effects were premature death or in those that survived, partial sterility (as low as a single litter) or complete sterility (Table I).

TABLE I

Animals studied	Total No. of Animals	Longevity upto		Mortality before maturity	
		14 Months	15 Months and more	Young	Dead
C57 Control	45	8 (17·7%)	37 (82·2%)	..	..
C57 Treated F	35	20 (60%)	15 (40%)	..	..
F_1	36	24 (66·66%)	12 (33·33%)	110	(68%)
Swiss Control	40	6 (15%)	34 (85%)	..	..
Swiss Treated F	43	16 (37·29%)	27 (62·79%)		
F_1	126	37 (29·37%)	89 (70·63%)	144	(67·3%)

TABLE II

Animals studied	Total No. of breeders	No. of Animals bred	Total litters	Av. No. of litters	Total No. of young born	Litter size	Sterile animals
C57 Control	25	25 (100%)	151	6·00	1123	7·4	..
C57 Treated F	20	19	23	1·2	161	8·4	..
F_1	21	2	2	0·100	11	5·5	19 (90·5%)
Swiss Control	20	20	122	6·1	770	6·3	..
Swis Treated F	24	23	42	1·75	270	6·4	73 (60·5%)
F_1	61	24	43	1·8	277	6·4	

Normal litters born, were kept for mating continuously from puberty till old age. Even then the treated mice remained partly sterile and the most affected strain was C 57 (Table II). Sterility in the F_1 generation was very high and the average number of litters was significantly low as a result of DDVP spray (Table II).

Histology :

Histology of the testes of the treated males revealed the presence of large vacuoles in them as a result of the deletion of seminiferous tubules. Germ cells were lacking in many tubules and cryptorchism was a common feature of these animals. Testes tumours were seen in two of the treated animals. Ovaries of treated females were hyalinized in nature and the follicles and oocytes were absent in a large number of animals.

Discussion :

The deleterious effects of insectides on breeding performance of various animals have been observed by several workers[4-8]. According to Hodjat (1971)[7] shorter exposure to DDT at 6·20 mg/litre caused reduced egg production and fertility in *Dysdercus fasciatus*. Lineva (1962)[9] claimed that sublethal exposure to insecticide induced abnormalities in the oogenesis of house fly. In the present experiment, animals had only environmental contact with DDVP. However, it is significant that a large number of treated C 57 animals were sterile (Table II). The histological data of the testes and ovaries of treated animals explain the pathogenicity of this insecticide. The fall in the longevity of the pesticide exposed animals (Table I) might be due to the general toxicity of the chemical[10]. The data presented in this paper indicate that DDVP spray causes sterility in mouse by bringing about drastic abnormalities in the reproductive systems.

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1. Verret, M. J.. Mutchler, M. K., Seott, W. F., Reynaldo, E. F. and McLaughlin, J., *Ann.*. 1969.
2. Marlack, J. P., *Fed. Proc.*,)964, 23, 105.
3. Somers, L. E. and Hsu, T. C., *Proc. Natl. Acad. Sci.*, 1962, 48, 937.
4. Auerbach. C., *Mut. Res.*, 1967, 158, 1141.
5. Fahmy, O. G. and Fahmy, M. J., *Roy. Soc. Trop. Med. Hyg.*, 1964, 58, 318.
6. Coaker, T. H. and Smith, L. J., *Bull. Ent. Res.*, 1970, 60, 53.
7. Hodiat. S. H., *Ibid.*, 1971, 60, 367.
8. Pillai, M. K. K. and Grover, K. K., *Bull. Wld. Hlth. Org.*, 1969, 40. 229.
9. Lineva, V. A., *XI Int. Cong-Ent.*, 1962, 2, 448.
10. Wasserman, M., Wasserman, D., Gershon, Z. and Zellermaver, L., *Ann. N.Y. Acad. Sci.*, 1969, 160, 393.

CORONA STRUCTURES AND THE ORIGIN OF HORNBLENDE-BEARING ROCKS OF S. WAYANAD, KERALA STATE

In S. Wayanad (Calicut Dist.) hornblende-bearing gneisses containing amphibolite and minor schistose bands constitute the major rock types in contrast to charnockites in south and central Kerala. However, these hornblende-bearing rocks carry frequently, as in south and central Kerala, pyroxene granulites as thin disconnected patches and lenticular boudins, rarely extending beyond a few meters along the strike. The gneisses, often banded and occasionally streaky, and schists have developed a megascopic axial surface foliation, mainly after hornblende and biotite. The granulites though free from megascopic foliation were also involved in the deformation and metamorphism responsible for the production of the axial surface foliation[1].

Hornblende, developed on a large scale in the rocks of the present area containing hornblende-free pyroxene granulites, can be either a granulite facies prograde[2] or an amphibolite facies retrograde product, or both.

Features characteristic of granulite facies hornblende[3] such as greenish brown colour, presence of opaque granules along hornblende grain boundaries[2] and pyroxene coronas around hornblende and alteration of hornblende into pyroxenes are not seen. Since sufficient quartz is present, failure of hornblende break-down reactions cannot be ascribed to lack of quartz. Hence hornblende is not likely to be a prograde product.

On the other hand, some of the pyroxene granulite patches in the greisses around Chundale (11° 34′ 30″: 76° 3′ 30″) show coronas of secondary hornblende around pyroxenes (Fig. 1). Synentatic rims are seen around both ortho- and clino-pyroxenes. The formation and width of the rims are independent of the nature and grain size of the pyroxenes. As a rule, the corona rims are formed when pyroxene is in contact with plagioclase. However, such rims, though incomplete, are also seen along the contact of adjacent pyroxene grains, especially in the vicinity of plagioclase. The corona rim consists of a fine symplectite

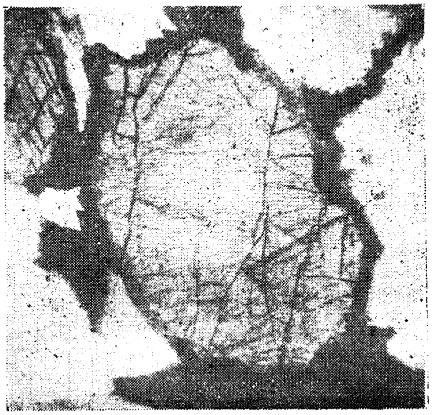

Fig. 1. Clino-pyroxene with corona. Lighter parts of the structure formed of intergrowth of hornblende and quartz and the darker parts of opaque ores. Plane polarised light, × 40.

intergrowth of short hornblende prisms perpendicular to the pyroxene grain boundary, irregular granules of opaque ores and minute crystals of quartz. The distribution of opaque ores in the rim is irregular; some portions show a higher concentration than others and some are even devoid of them. The width of one and the same rim may vary from place to place. In thicker portions the amount of hornblende increases progressively at the expense of opaque ores and in advanced stages opaque ores either completely disappear or are present only as occasional inclusions in hornblende. There seems to be a direct relation between the relative amounts of secondary hornblende and opaque ores in the corona rim and the degree of coronation. Since in the majority of cases coronas have formed only around pyroxene grains in contact with plagioclase felspar, an igneous origin is precluded. Hornblende coronas around pyroxenes, representing frozen stages of reaction between adjacent pyroxene and plagioclase mineral phases, can form according to the following reaction :

$$\text{pyroxene} + \text{plagioclase} + \text{water} \rightleftharpoons \text{hornblende} + \text{quartz.}$$

If water in required amounts is not available during superposed amphibolite facies metamorphism of granulite facies rocks, break-down and conversion of pyroxenes into hydrous minerals like hornblende and biotite cannot take place. This might explain the preservation of water-deficient basic granulites as relics in the hornblende-bearing rocks and rarity of coronas in the relict granulites of the area. As only small amounts of water are necessary for corona formation[4], its availability in such amounts locally[5] in the granulites must have favoured the formation of local hornblende rims around pyroxene grains.

The hornblende gneisses and amphibolites are almost free from pyroxenes. The pyroxene grains when rarely encountered are invariably clouded and do not show hornblende rims. This suggests that pre-existing pyroxene grains have mostly been converted completely into hornblende during retrograde amphibolite facies metamorphism. Further, this must have happened under sufficient or excess water conditions since such conditions are believed to favour reactions outside mineral individuals and destroy corona relations[6].

In addition, the gneisses and amphibolites show the following features characteristic of the development of amphibolite facies rocks from a granulite facies parent[3].

1. Hornblende in the gneisses and amphibolites of the area is the bluish green to grass green variety in contrast to the characteristic deep greenish brown and brown variety found in the granulite facies rocks of S. Kerala and the neighbouring Kanyakumari District of Tamil Nadu.
2. The rocks have developed gneissic structure and foliation after hydrous minerals, *viz.*, hornblende and biotite.
3. Hornblende-quartz symplekite intergrowths are common in the gneisses and amphibolites.
4. The rocks contain microcline, and the perthite present is the mesoperthite variety.

No direct evidence regarding the nature of the parent rock from which the hornblende-bearing rocks were derived during amphibolite facies metamorphism is observed. However, it is significant that charnockites, the common rock types in many parts of Kerala, are extremely rare in the area. This, together with the evidences presented above, points to the possible development of hornblende-bearing rocks mainly at the expense of charnockites through retrograde amphibolite facies metamorphism. A similar polymetamorphic origin has been suggested for the Pre-cambrian Vijayan Series rocks of Ceylon[7].

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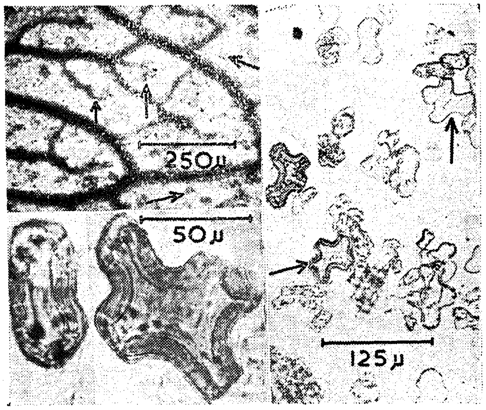
1. Nair, P. K. R. and Michael, G. P., *Curr. Sci.*, 1975, **44**, 230.
2. Sen, A. K. and Sobhen Ray, N., *Jb. Miner. Abh.*, 1971, **115**, 291.
3. Katz, M. B., *Geol. Mag.*, 1968, **105**, 487.
4. Murthy, M. V. N., *Bull. Goel. Soc. Amer.*, 1958, **63**, 34.
5. Griffin, W. L. and Heier, K. S., *Contr. Mineral. and Petrol.*, 1969, **23**, 89.
6. Bose, M. K., *Geol. Mag.*, 1961, **98**, 415.
7. Cooray, P. G., *Quar. Geol. Soc. Lon.*, 1962, **118**, 255.
8. Spry, A., *Metamorphic Textures*. Pergamon Press, 1969.

FOLIAR SCLEREIDS IN *MICROTROPIS VALIDA*

THE presence of sclereids (Sclerenchymatous idioblasts) is accounted in the mesophyll tissues of *Maurocenia*, *Maytenus*, *Microtropis*, *Pterocelastrus* and *Schaefferia* of Celastraceae[1]. Further details regarding their structure, distribution, density and relationship within the mesophyll are not known. In the two other genera namely *Lophopetalum* and *Kokoona* brachysclereids are reported from leaf tissues[2]. Vesiculose sclereids are present in leaves of *Goupia glabra*[3] and this taxon has been shifted into a new family Goupiaceae. The observations made on the foliar sclereids in *Microtropis valida* Ridl. are briefly reported here.

Microtropis is an Indo-Malayan genus with about 13 species restricted in distribution to South Asia. Five species, including *M. valida*, are endemic to Malaysia and Borneo[4]. *M. valida* is a small tree growing up to 5–7 m, with thick, opposite leaves. Fresh leaves were collected from the trees growing in Kota Tinggi area, South Johore, West Malaysia. Customary methods were followed to obtain leaf clearings, sections and macerations.

In leaf clearings 90% of the sclereids show diffuse distribution. Since the leaf is thick ($480\,\mu$), epidermal surfaces are covered by thick cuticle, and sclerieds are confined to the middle spongy layers, the sclereid distribution is somewhat hazily seen in leaf clearings (Fig. 1). Comparatively they are more clearly seen from the lower epidermis side than the upper. Regarding terminal sclereids, either they are solitary or occur in groups of 2–3 at vein endings (Fig. 1).



FIGS. 1–4. Fig. 1. Leaf clearing showing sclereid distribution (vertical arrows—terminal sclereids, horizontal arrows—diffuse sclereids). Figs. 2, 3. Brachy- and vesiculose sclereids enlarged to show shape and cell wall thickness. Fig. 4. Part of macerated tissue showing similarity in shape between spongy mesophyll cells and vesiculose sclereids (arrows).

In macerated preparations two types of sclereid forms are seen; the first type of spheroidal or oval-shaped brachysclereids and the second type of vesiculose sclereids. The former are unbranched whereas sclereids of vesiculose type show 3–4 lobes, some of them with flattened, enlarged ends, or each lobe branched further with two flat edges (Figs. 2, 3). Both the types of sclereids are primarily oriented parallelly to the surface of the lamina.

The transverse sections of the lamina reveal that the sclereids are mostly confined to spongy mesophyll tissue, most of them being present in the middle layers.

In the macerated preparations the close similarity in cell shape between the sclereids and spongy mesophyll cells is clearly seen (Fig. 4). The bigger spongy mesophyll cells are lobed, each lobe with a curved or dome-shaped outline. In distinction from the spongy cells the sclereid cells have enlarged 2–3 times, with flat or branched cell lobes. But the sclereids do not radically deviate in shape from the spongy mesophyll cells (Fig. 4). The oval-shaped (length $45\,\mu \times$ width $20\,\mu$) sclereids are smaller than the vesiculose sclereids ($63\,\mu \times 42\,\mu$), and these are the average figures obtained from 20 measurements in each case. The cell wall is thick ($6\,\mu$), lamellated, with many simple pits, evenly spaced. In some, the wide lumen consists of degenerated nucleus and cytoplasm.

The constancy of cell shape has been considered as an important criterion in classifying sclereids under monomorphic and polymorphic types[5,6]. The range of variation seen under monomorphic types is limited and the two types of sclereids in *M. valida* are of monomorphic type. Additional evidence from sclereid characters has been recently produced lending support for the recognition of a separate family, Goupiaceae[3]. Genus *Goupia* was originally included in Celastraceae. In view of this, detailed studies on sclereids are necessary in different genera of this family. A preliminary survey made on other species of *Microtropis* has shown that foliar sclereids are absent in *M. bivalvis*, *M. curtisii*, *M. discolor*, *M. kinabaluensis*, *M. petelotii*, *M. platyphylla*, *M. sumatrana*, *M. tetrameris* and present in *M. elliptica*, *M. fukinensis*, *M. osmathoides* and *M. wallichiana* (= *ramiflora*). Details of leaf anatomy and sclereid characters in this genus will be published elsewhere.

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Singapore, *September* 27, 1975.

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1. Metcalfe, C. R. and Chalk, L., *Anatomy of the Dicotyledons*, Clarendon Press, Oxford, 1950.
2. Janser, W. T. and Baas, P., *Blumea*, 1973, **21**, 153.
3. Rao, T. A. and Bhattacharya, J., *Curr. Sci.*, 1975, **44**, 132.
4. Ding Hou, *Celastraceae*, *Flora Malesiana*, Walters-Noordhoft Publ., Groningen, 1972.
5. Rao T. A. and Bhupal, O. P., *Proc. Indian Acad. Sci.*, 1973, **77**, 41.
6. De Roon, A. C., *Acta Bot. Neerl.*, 1967, **15**, 585.

A NEW RUST ON *POLYGONUM GLABRUM* FROM INDIA

DURING mycological survey of Khandala (near Lonavala) rust infected leaves of *Polygonum glabrum* L. (Polygonaceae) were collected. The rust was found to belong to the uredinial stage, when studied microscopically. The present uredinial form was found to be in conformity with uredinial genus *Uredo* Pers. amongst several uredinial forms[1]. Further comparative studies showed that the present species could not be accommodated in any of the previously described species of *Uredo* affecting the members of Polygonaceae[2,3], chiefly on account of smaller spore size, position of germ pore (all supra-equatorial), thicker spore wall, cushion-shaped pedicel attachment and unrecorded host species. The present material, therefore, is placed under a new specific taxon and the necessary Latin diagnosis is given below.

Uredo khandalensis sp. nov. (Plate I).

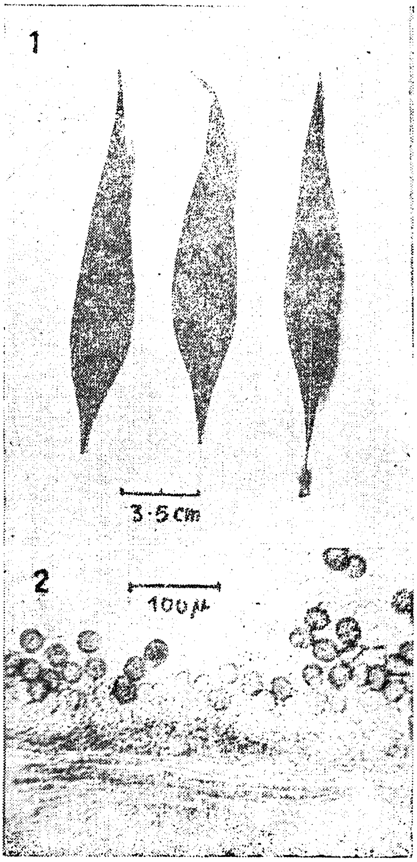

PLATE I. *Uredo khandalensis* sp. nov. 1. Infected leaf. 2. Urediniospores.

Uredinia plerumque abaxialia, cinnamoneo-bruneia, rotundatia, aparaphysalia, magnit 0·5–0·9 mm, urediniosporae ellipsoideae variabiles, sphericae vel pyriformes complanatum versus hilum, cinnamoneo-bruneiae, echinulatae, magnit 18·0–23 (–29) × (14–) 16–19 μ; pariete 3–4 μ crassa, 4 μ crassa ad hilum; hilum pulvinatum; pedicelius hyalinus, magnit 44–46 × 4–6 μ latus ad apiceum; germ pori 2 (raro 3), supraequatorii.

Teleutosporae non-cognitae.

In foliis viventibus *Polygonae glabrae* L.

Holotypus AMH* subnumero 2567.

Typus locus Khandala (M.S.) India.

Authors are grateful to Prof. M. N. Kamat, Head, Department of Mycology and Plant Pathology, for his interest. One of us (S. R. R.) is grateful to Dr. G. B. Deodikar, Director, M.A.C.S., for the award of Junior Research Fellowship. The help rendered by Dr. (Mrs.) A. Pande in translating the diagnosis in Latin is duly acknowledged.

Maharashtra Association A. V. SATHE.
for the Cultivation of Science, S. R. RAHALKAR.
Poona 411 004,
September 22, 1975.

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* AMH—Ajrekar Mycological Herbarium at M.A.C.S., Poona-4.

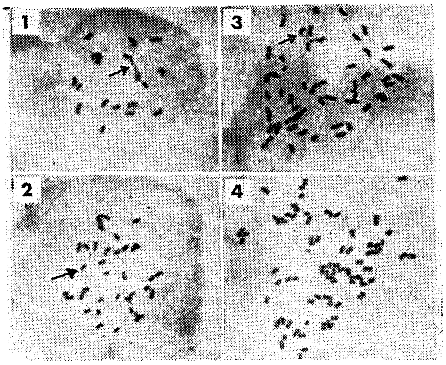
1. Sathe, A. V., *Origin and Evolution of Uredinia in Rust Fungi*, Part of Ph.D. Thesis, Poona University (Unpublished), 1969.
2. Savulescu, T., *Monografia Uredinalelor, Part II.* Academic Republic Popular Romania Bukharest, 1953, p. 1166.
3. Gaumann, E., *Die Rostpilze Mitteleuropas*, Buchdruckerei, Buchler and Co., Bern., 1959, p. 1407.

SQUASH METHOD FOR THE MITOTIC CHROMOSOMES OF GRASSES

THE grasses in general are difficult materials for mitotic studies. The factors responsible for this are firstly, the excessive hardness of their root-meristems which does not permit sufficient softening of the cells and consequently proper spreading and spacing between the chromosomes; and secondly, the extremely small size of their chromosomes (often large in numbers) which makes it hard to stain them and to achieve a desirable contrast between the cell-cytoplasm and chromosomes. It is for these reasons that some distinguished grass cytotaxonomists have exclusively depended on meiotic chromosomes in their studies (Gould, 1966; Gould and Soderstrom, 1967), while others have been making use of microtome sections for determining the somatic numbers of chromosomes in grasses (Tateoka, 1965 and others).

The "Feulgen" method (Darlington and La Cour, 1962) as well as several modifications of the aceto-carmine method (Howe, 1946; Smith, 1947 and others) for staining root-tip chromosomes of grasses do not give satisfactory results. Two modifications of the aceto-carmine method have been specifically recommended for grasses. Hensen and Oldemeyer (1951) recommend aceto-carmine to reach boiling point while the roots are being stained. Powell (1968) recommends pectinase digestion of the leaf tissue of forage grasses prior to staining in aceto-carmine or aceto-orcein. Apparently the main purpose of both of these modifications is to soften the meristematic tissue.

The following schedule which makes use of hydrochloric acid of a high concentration for separating and softening the root-meristem cells of grasses followed by staining of their chromosomes in Heidenhain's haematoxylin has given us excellent results in a large number of grasses. The results obtained by this method are illustrated by Figs. 1–4, obtained from four species of *Digitaria* forming a polyploid series.



FIGS. 1–4. Mitotic metaphases in root-tip cells of four *Digitaria* species. Fig. 1. *D. longiflora* (Retz) Pers, $2n = 18$. Fig. 2. *D. ischaemum* Scherb, $2n = 36$. Fig. 3. *D. adscendens* (HBK) Henr, $2n = 54$. Fig. 4. *D. griffithii* (Hook. f.) Henr, $2n = 72$.

The arrows point to the satellited chromosomes. All photomicrographs *c.* × 925.

The roots are treated in saturated aqueous solution of α-Bromonapthalin, for half an hour prior to fixation in 1 : 3 acetic alcohol for 10–15 minutes. The roots are then softened in 50% HCl for about 45 minutes at room temperature. This time will vary according to the degree of hardness of the material. After thorough washing in tap water they are mordanted in 2% iron alum solution for 5 minutes. They are then washed thoroughlly in tap water to remove all traces of iron alum and finally stained in 0·5% aqueous solution of Haematoxylin for 4–6 minutes after which the roots are rinsed in tap water. They are now ready for making squash preparation in 0·5% aceto-carmine. The preparations are made permanent in the usual way using ethyl alcohol and euperol.

In addition to many grass species we have tried this method with excellent results in a variety of other materials containing small chromosomes in varying numbers, such as, sweet potato (*Ipomoea batatas*), $2n = 90$; groundnut (*Arachis hypogaea*), $2n = 40$; *Sesbania* spp., $2n = 12$ and 24; several cultivated and wild jute species (*Corchorus*) $2n = 14$ and 28; tomato (*Lycopersicon esculentum*), $2n = 24$, etc. In every case, it has been possible to observe the details of the karyotype, such as, the length of the chromosomes, the positions of the centromere, the length of the two arms of the chromosomes and the positions of the secondary constriction of the satellited chromosomes.

It is to be mentioned that in materials other than grasses where the meristematic cells are not hard, the duration of the HCl treatment is to be considerably reduced.

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Mymensingh,
Bangladesh, *July* 5, 1975.

ASHRAFUL HAQUE.
M. ARSHAD ALI.
MD. WAZUDDIN.
MD. A. KARIM.

1. Darlington, C. D. and La Cour, L. F., Allen and Unwin, Ltd., London, 1962.
2. Gould, F. W., *Canad. Jour. Bot.*, 1966, **44**, 1683.
3. — and Soderstrom, T. R., *Amer. Jour. Bot.*, 1967, **54** (6), 676.
4. Hanson, A. A. and Oldemeyer, D. L., *Stain Tech.*, 1951, **26**, 241.
5. Howe, J., *Ibid.*, 1946, **21**, 127.
6. Smith, L., *Ibid.*, 1947, **22**, 17.
7. Powell, J. B., *Ibid.*, 1968, **43**, 135.
8. Tateoka, T., *Amer. Jour. Bot.*, 1965, **52** (8), 864.

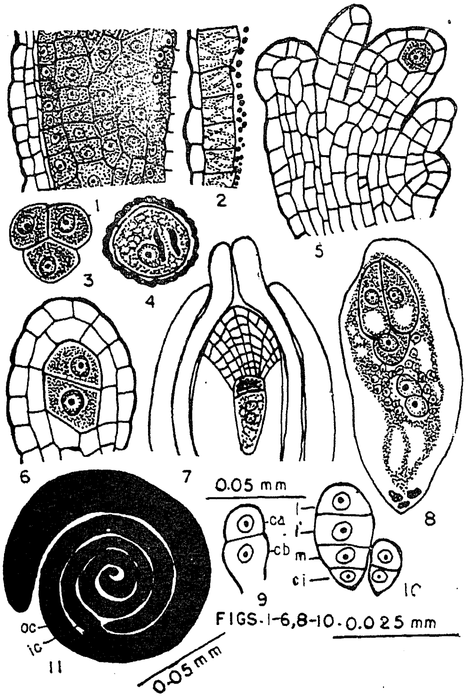
EMBRYOLOGY OF *SUAEDA MARITIMA* (LINN.) DUMORT.

THE genus *Suaeda*, a member of Chenopodiaceae, is a halophyte and comprises 110 species (Willis, 1966). A few of them occur both in east and west coasts of India. Although some embryological work has been done on the species of *Suaeda* (Mahabale and Solanky, 1953) occurring in the west coast, the species occurring in the east coast remain uninvestigated. The present account deals with the life-history of *S. maritima* (Linn.) Dumort.

The anther is tetrasporangiate and its wall development conforms to the monocotyledonous type (Davis, 1966). The glandular tapetum is uniseriate and consists of one or two nucleate cells (Fig. 1). The middle layer is ephemeral. The subepidermal layer develops into the fibrous endothecium. On the inner surface of the fibrous layer small, globular structures are present (Fig. 2) as in *S. fruticosa* (Mahabale and Solanky, 1953). In the mature anther, almost all cells of the anther except a few connective cells, epidermis and fibrous layer, become degenerated. The epidermal cells at the region of the dehiscence become bulliform and aid in the dehiscence of the anther. The pollen mother cells undergo simultaneous cytokinesis and produce pollen tetrads which are either tetrahedral (Fig. 3) or isobilateral. The pollen grains are three-celled (Fig. 4) and multiaperturate at the time of shedding. Degeneration of one of the pollen sacs of the anther is a common feature.

The ovary is superior, bi- or tricarpellary syncarpous and unilocular with a single basal ovule. The ovule is campylotropous, bitegmic and crassinucellate. The inner integument forms the micropyle. A prominent air space is present at the base in between the two integuments. The archesporium is single celled (Fig. 5). It divides periclinally and produces a primary parietal cell towards outside and a sporogenous cell towards inside which develops into the megaspore mother cell. A well-developed parietal tissue is formed (Fig. 7). The megaspore mother cell undergoes the first meiotic division followed by wall formation and produces dyad of cells (Fig. 6). The upper one degenerates and the lower dyad after undergoing three more divisions develops into an eight nucleate embryo sac of the Allium type (Figs. 7, 8). Fertilisation is porogamous. Syngamy and triple fusion occur more or less simultaneously. Endosperm is *ab initio* Nuclear. Cell wall formation commences from the micropylar region and proceeds towards the chalazal region and ultimately filling the entire embryo sac with cellular tissue. The endosperm is completely consumed by the developing embryo. The zygote divides transversely and produces two celled embryo (Fig. 9). The terminal cell *ca* and the basal cell *cb* undergo one more transverse division each and produce a four celled linear proembryo. The derivatives of *ca* are termed as *l* and *l'* and those of *cb* as *m* and *ci*. The derivatives of both the terminal cell *ca* and the basal cell *cb* contribute to the formation of the embryo proper and thus the embryogeny in *S. maritima* conforms to the Chenopodiad type. The mature embryo becomes spirally coiled. The radicle, hypocotyledonary region and cotyledons are very much elongated. The inner cotyledon is larger than the outer one (Fig. 11). The cells of the mature embryo contain abundant food materials.

In a few cases, however, polyembryony is observed (Fig. 10). Judging by the position it seems that the second embryo might have originated from the synergid.



FIGS. 1–11. *Suaeda maritima.* Fig. 1. L.s. of anther lobe showing epidermis, wall layer, tapetum and sporogenous tissue. Fig. 2. Fibrous endothecium and globular bodies. Fig. 3. Microspore tetrad. Fig. 4. 3-celled pollen grain. Fig. 5. L.s. ovule. Fig. 6. L.s. nucellus showing dyad. Fig. 7. L.s. upper part of ovule showing integuments, parietal tissue and functional dyad. Fig. 8. Eight nucleate embryo sac. Fig. 9. 2-celled embryo. Fig. 10. Twin embryos. Fig. 11. Coiled embryo. (*oc*, outer cotyledon ; *ic*, inner cotylendon.)

In a mature seed the cells of the inner layer of the inner integument and outer layer of the outer integument radially elongate, develop tannin contents and finally develop into the seed coat.

The two investigated species of *Suaeda*—*S. fruticosa* and *S. maritima* (Mahabale and Solanky, 1953 and present data)—show great uniformity in their embryological characters like the development of the anther, male gametophyte, ovule, female gametophyte, endosperm, embryo and seed coat. However, polyembryony present in a few cases of *S. maritima* and it is absent in *S. fruticosa*. The genus *Suaeda*, although resembles the rest of the Chenopodiaceae in the majority of embryological characters, it differs from them in certain characters and a detailed discussion of which will be presented elsewhere.

The junior author is thankful to the authorities of Andhra University for the award of University research fellowship.

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Waltair, *September* 30, 1975.

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1. Davis, G. L., *Systematic Embryology of Angiosperms,* John Wiley and Sons, Inc., New York, 1966.
2. Mahabale, T. S. and Solanky, I. N., *J. Univ. Bombay,* 1953, 21, 81.
3. Willis, J. C., *A Dictionary of the Flowering Plants and Ferns,* Cambridge, 1966.

TWO *ASCOBOLUS* SPECIES FROM INDIA

DURING a study on Indian Discomycetes the author came across two species of the genus *Ascobolus* Pers. ex Fr., *viz., A. brassicae* Crouan and *A. behnitziensis* Kirschst., which were collected during 1967–68, and have not been reported before from this country. Concise descriptions of these species are provided in the present note. Specimens studied have been deposited in Banaras Hindu University, Plant Pathology Herbarium, abbreviated as BHUPP.

Ascobolus brassicae Crouan, *Annls. Sci. nat.* (*Bot.*) IV 7, 174, 1857.

(Figs. 1–3)

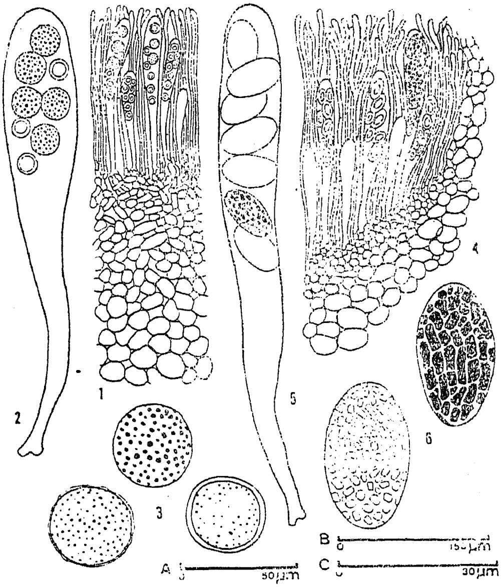
Apothecia scaltered to gregarious 1–2, mm in diam., sessile, closed at first, later becoming slightly obconical to scutellate ; hymenium olivaceous brown ; external surface lighter than the hymenium ; margin reddish brown, crenate. In section : hymenium 180–220 µm thick ; subhymenium and medullary excipulum not distinguishable separately, together 100–180 µm thick, consisting of textura intricata, hyphae up to 18 µm wide, at places forming swollen cells ; ectal excipulum 40–100 µm thick, consisting of textura angularis or text, globulosa, cells up to 25 µm in diam ; asci faintly blue in Melzer's reagent when young otherwise pseudoamyloid, 8-spored, only 1–5 spores attaining maturity, clavate with somewhat conical apex, narrowing below into short bases, 125–175 × 16–25 µm ; ascospores hyaline, globose to completely spherical, purple to light yellow brown, eguttulate, marked with small, isolated, round or angular warts, uniseriate when young, later irregularly arranged, 13–16·5 µm in diam. paraphyses hyaline, filiform, septate, 1–2 times branched at the lower portion, up to 3 µm wide at their apices.

Specimen studied : BHUPP 680, on soil, behind Banaras Hindu University Press, Leg. K. B. Khare, January 27, 1968.

A. brassicae Cr. has been reported on dung of various animals and on rotten stems of Cabbage[1]. Soil is being reported here an additional substrate for this species.

Ascobolus behnitziensis Kirschst., *Verh. bot. Ver. Brandenburg,* 48, 47, 1907.

(Figs. 4–6)



FIGS. 1–6. Figs. 1–3. *Ascobolus brassicae.* Fig. 1. Part of excipular tissue and hymenium. Fig. 2. A complete ascus. Fig. 3. Ascospores. Figs. 4–6. *Ascobolus behnitziensis.* Fig. 4. V.S. of an apothecium showing excipular tissue and hymenium. Fig. 5. A complete ascus. Fig. 6. Ascospores. Scale line : A for Figs. 2 and 5 ; B for Figs. 1 and 4 ; C for Figs. 3 and 6.

Apothecia scattered, up to 5 mm in diam. seccile, shallow cupulate to scutellate or flat ; hymenium olive green, black dotted due to protruding tips of ripe asci ; externally olive brown, smooth. In section : hymenium 190–255 µm thick ; subhymenium 30–62 µm thick, of short, isodiametric, angular cells, cells 2–5 µm in diam. ; medullary and ectal excipulum not distinguishable separately, together 50–250 µm thick, consisting of textura glubulosa, cells 10–30 µm in diam., larger towards outside and smaller in the middle of the eycipulum ; asci non-amyloid (walls blue in Melzer's reagent when young), 8-spored (in few asci only 4–6 spores attaining maturity), cylindric-clavate, narrowing below, 180–250 × 20–32 µm ; ascospores hyaline, elliptic, narrowing slightly towards ends, violet to dark brown with yellow tinge, ornamented with a network of crevices or striae which are dissolved in 2% KOH, uniseriate when young, later biseriate or irregularly arranged, 22–32 × 12–20 μm; paraphyses simple to branched, septate, embedded in yellow mucus.

Specimen studied: BHUPP 439, on soil, Pantnagar—Nainital road side, Leg. D.C. Pant, October 30, 1967.

Asci and ascospores, in the present specimen, are slightly larger than that described by Brummelen[1] for this species.

I am thankful to Prof. M. S. Pavgi for providing laboratory facilities and to Shri V. P. Tewari, for the encouragement. The help of C.S.I.R., New Delhi, for appointing me in the Scientist's Pool is gratefully acknowledged.

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October 3, 1975.

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1. Brummelen, J. Van, *Persoonia* (*Suppl.*), 1967, 1, 1.

EPIDERMAL STRUCTURE AND DISTRIBUTION OF STOMATA IN *APONOGETON NATANS* (L.) ENGL. AND KRAUSE

Aponogeton natans is a freshwater, floating aquatic herb of the family Aponogetonaceae which occurs rather commonly in the ponds of Northern India. The author observed a few plants growing in the mud outside the pond wherefrom the water had receded. These plants were completely aerial with their tuberous rhizomes buried in the soil. The present work was undertaken to study the distribution of stomata on the two surfaces of the aerial, floating, and submerged leaves and their correlation with the habitats to which they were exposed.

The material was fixed in FAA and subsequently stored in 70% alcohol. The epidermal peels from the upper and lower surfaces of aerial, floating and submerged leaves were taken out, stained with Delafield's hematoxylin and mounted in glycerine jelly.

In their gross morphology the aerial and floating leaves tend to be simple with a long, slender petiole. The latter usually terminates by a shallow, cordate base of the leaf. The submerged leaves on the other hand are linear lanceolate.

The aerial leaves possess polygonal epidermal cells with slightly curved walls—those of the lower epidermis being larger than the ones composing the upper tepidermis. The upper epidermal cells of the floaiing leaves on the other hand are elongated and smaller as compared to their counterparts on the lower surface, which are also polygonal in outline. Both the upper and lower epidermal cells in aerial leaves are smaller than those of the floating and submerged ones (Table I).

TABLE I

Frequency of epidermal cells and stomata on the upper and lower surfaces of leaves of A. natans *under various habitats*

Habitat	Surface	No. of epidermal cells per sq. mm.	Stomatal frequency per sq. mm.
Aerial	upper	2107	183
	lower	1212	21
Floating	upper	1761	179
	lower	1156	13
Submerged	upper	1740	21
	lower	1045	10

The aerial and floating leaves are amphistomatic but the frequency of stomata on the lower surface is much lower as compared to the upper (Table I). The stomata are oriented parallel to the veins. However, it is interesting to note that the stomata on the upper epidermis of the floating leaves are more regular in orientation and larger than those on the upper surface of the aerial leaves.

The cubmerged leaves possess stomata only on the upper surface. Even here the stomatal frequency is very low. Moreover, they are smaller in size as compared to those on the upper surface of aerial and floating leaves. The various types of stomatal complexes have been given in Table II following Dilcher[1].

TABLE II

Types of stomatal complex

Leaf types	Upper surface	Lower surface
Aerial	Brachyparacytic	Atinocytic
Floating	do.	do.
Submerged	do.	Stomatal complex absent

In the floating leaves the distribution of stomata is variable. Generally they are restricted entirely to the upper epidermis; the lower having only a few of them. This is perhaps in agreement with the observations made by Gupta *et al.*[2] on *Nelumbo nucifera.* According to these authors the emerging, terrestrial leaves of this plant have a few stomata on the lower surface as well (being much fewer than those present on the upper surface). These slowly get disorgtnized and are finally lost as the area gets flooded and the plant comes to acquire the floating habit. Thus at maturity, the upper epidermis alone is provided with stomata.

Shinobu[3] also reported the occurrence of stomata on the lower epidermis of the floating leaves of two species of *Potamogeton,* viz., *P. fryeri* and *P. gramineus.* He found a mean stomatal frequency of 3 and 4 per sq. mm. respectively on the lower epidermis of *P. fryeri* and *P. gramineus* as compared to the values of 136 and 147 per sq. mm for the upper surface. He also recorded a difference in the occurrence of stomata on the lower epidermis of these plants according to their habitat.

In the present study, although the author did come across stomata on the lower epidermis of the floating leaves of *A. natans,* yet their frequency was much lower than that on the upper surface as recorded for the species of *Potamogeton.* However, their number per unit area is much higher to the figures recorded by him[3] (*see* Table I). Moreover the submerged leaves also possess stomata on the upper surface although they are absent from the lower epidermis.

Thus as regards the distribution of stomata on the lower surface of the floating leaves of aquatic plants we have three situations. To the first category belong the leaves of *Nelumbo nucifera* where the mature leaves are completely devoid of stomatal apparatus (Gupta *et al.*[2]); those of *Potamogeton* species have only a few of them (Shinobu[3]) and finally the leaves of *Aponogeton natans* which have reasonably higher number of stomata distributed even on the lower surface (present work).

According to Shinobu[3] the stomata in the lower epidermis of floating leaves are functionless and are relic ancestral features and so are the stomata occurring on the upper surface of the submerged leaves. This supports the idea of Porsch[4] that the stomata are complex structures which once acquired, irrespective of their utility, appear to become hereditarily associated with the plant organ.

These findings also have agreement with Haberlandt's[5] preposition that stomata in aquatic plants reflect an ancestral relationship with terrestrial plants.

I am grateful to the authorities of the college for granting me permission to carry out this study and for providing necessary facilities.

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December 20, 1975.

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1. Dilcher, D. L., *Bot. Rev.*, 1974, 40, 1.
2. Gupta, S. C., Paliwal, G. S. and Rani Ahuja, *Amer. J. Bot.*, 1968, 55, 295.
3. Shinobu, R., *Bot. Mag. Tokyo,* 1952, 65, 56.
4. Porsch, O., "Der Spaltöffnungsapparat", In: *Lichte der, Phylogenie,* Jena, 1905.
5. Haberlandt, G., *Physiological Plant Anatomy,* English Translation, Macmillan and Co. Ltd., London, 1914.

MICROSPOROGENESIS AND MALE GAMETOPHYTE IN *RAUWOLFIA SERPENTINA* (L.) BENTH. EX KURZ

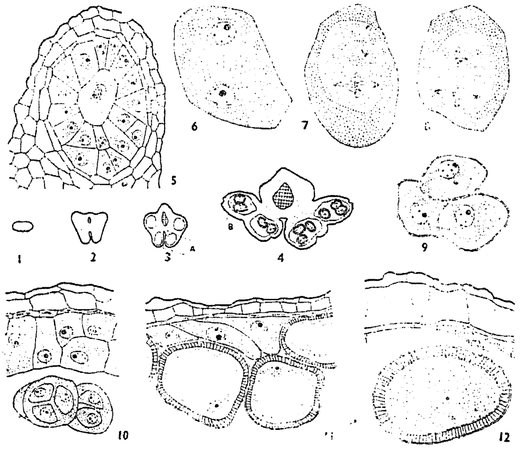
Rauwolfia serpentina is an important medicinal plant belonging to Apocynaceae. No information existed on the embryology of the species till recently when Lamba (1974)[1] reported its megasporogenesis and female gametophyte. Earlier, Meyer (1938)[2] investigated the development of pollen and embryo sac in *Rauwolfia canescens.*

The material for the present work was collected round about Delhi. The flower buds were fixed in F.A.A. and usual embryological procedure was followed.

In transection the young microsporangium has a circular outline and is composed of homogeneous parenchymatous cells. Gradually it becomes slightly oval and 2-lobed and finally 4-lobed (Figs. 1–2). The wall of the quadrisporangiate anther consists of an epidermis, an endothecium, a middle layer and the tapetum (Figs. 3–5). The tapetal cells remain uninucleate throughout and some of them may undergo a periclinal division resulting in a 2-layered condition (Figs. 5–10). Their dense cytoplasm becomes vacuolate by the time tetrads are formed (Fig. 10). The glandular tapetum degenerates only after the pollen reaches the 2-celled stage (Fig. 12). However, Frye and Blodget (1905)[3] and Anantaswamy Rau (1940)[4] observed its collapse in some Apocynaceae soon after the separation of microspores.

As microspore mother cells prepare for meiosis (Figs. 6–8), their protoplasts recede from the original wall and a special mucilaginous wall fills this space. Divisional stages ranging from early prophase I to late telophase II may be noticed in the microsporocytes contained in the same microsporangium. During meiosis II the spindles may be oriented parallel or at right angles to each other (Figs. 7–8) forming isobilateral, tetrahedral (Figs. 9–10) and decussate tetrads. As the young microspores enlarge the special mucilaginous wall is consumed and the original wall of the mother cell breaks down (Fig. 9). In *Rauwolfia canescens,* Meyer (1938)[2] noted both successive and simultaneous divisions. However, in *R. serpentina,* the author observed only simultaneous divisions.

Due to the appearance of a vacuole the centrally situated nucleus of the richly cytoplasmic microspore is soon pushed to one side (Fig. 11). Subsequent to mitotic division in the microspore, the generetive nucleus surrounded by a cytoplasmic sheath moves nearer the vegetative nucleus (Fig. 12). The mature pollen grain contains abundant starch (Fig. 12). The enlargement of the pollen grain is accompanied by the thinning of the exine. The pollen is shed at the 2-celled stage (Figs. 4–12) which is also the case in *Amsonia salicifolia* (Schnarf, 1931)[5]. Meyer (1938)[2] reported 3-celled condition in *Rauwolfia canescens* and some other Apocynaceae.



FIGS. 1–12.

In a mature anther the outer tangential wall of the epidermis becomes somewhat papillate. The cells of the endothecium elongate radially and develop fibrous thickenings (Fig. 12). The middle layers get flattened even before the formation of tetrads. The dehiscence of the anther is brought about by the breaking down of the thin walled cells at the junction of the pollen sacs (Fig. 4).

The author is highly grateful to Professor B. M. Johri for his valuable guidance, constant help and encouragement.

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Kurukshetra,
August 31, 1975/*September* 18, 1975.

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1. Lamba, L. C., *Acta Botanica Indica,* 1974, **2,** 154.
2. Meyer, Sally., *J. Department Sci.,* Calcutta Univ., 1938, **1,** 131.
3. Frye, T. C. and Blodget, E. B., *Bot. Gaz.,* 1905, **11,** 49.
4. Anantaswamy Rau, M., *J. Indian bot. Soc.,* 1940, **19,** 33.
5. Schnarf, K., *Vergleichende Embryologie der Angiospermen,* Berlin, 1931.

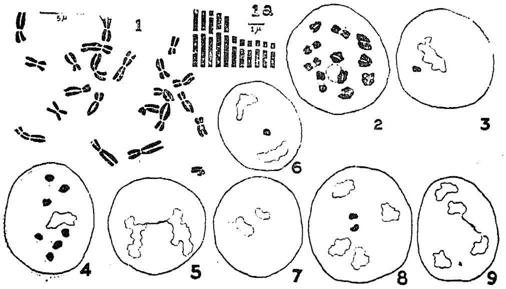
CYTOLOGY OF PONGAM OIL TREE [*DERRIS INDICA* (LAMK.) BENNET]

Derris indica (Lamk.) Bennet[2] [Syns. *Pongamia pinnata* (Linn.) Pierre., *Pongamia glabra* Vent.] is an economically important tree growing widely upto an altitude of 1200 meters (S. L.) in tropical Asia. Pongam oil from the seeds has many medicinal and industrial uses. The reports of only the chromosome number for the species are $2n = 20$[1] and $n = 11$ and $2n = 22$[3,4]. This communication deals with the karyotype and meiotic studies of the plant.

The seeds and flower buds were collected from different localities in Gujarat State. Following Tjio and Levan's (1950)[6] oxyquinoline aceto-orcein squash technique, the mitotic preparations were made. The flower buds were fixed in Carnoy's fluid (6 : 3 : 1) before preparing the smears. The pollen fertility was determined by using Muntzing's mixture[5].

The present investigation confirms the earlier reports of $n = 11$, $2n = 22$[3,4]. The chromosomes within the complement can be classified into 2 categories, *viz.,* the longer and the shorter types based on the length and ratio of the longest to the shortest pair of chromosomes. There are 5 pairs of long chromosomes ($3 \cdot 06\ \mu$ to $3 \cdot 90\ \mu$), of which 3 pairs are with nearly median and 2 pairs are with nearly sub-median centromeres. One of the 2 pairs with nearly median centromere is having a secondary constriction on the short arm. The short type of chromosomes is represented by 6 pairs ($1 \cdot 70\ \mu$ to $2 \cdot 55\ \mu$) having nearly sub-median centromeres without any secondary constriction (Figs. 1 and 1 *a*).

Meiotic studies show the presence of 11 bivalents (Fig. 2). Frequent occurrence of abnormalities were encountered during the first and second meiotic divisions. The common abnormalities recorded were occurrences of laggards, bridges, unequal distribution and cytomixes (Figs. 3 to 8).



FIGS. 1–9. Fig. 1 and 1 *a*. Karyotype and idiogram. Fig. 2. Normal diakinesis. Fig. 3. PMC showing association of 10 bivalents and 1 separate. Fig. 4. PMC showing association of 4 bivalents in the middle and 7 bivalents around. Figs. 5. and 6. Laggard and bridge at anaphase I. Fig. 7. Unequal distribution. Figs. 8. and 9. Laggards and bridge at anaphase II respectively.

From the total number 385 pollen mother cells scored for observations nearly 41·57% showed abnormal divisions. This behaviour was confirmed by determining the pollen fertility, which turned out to be 56·40%. The presence of abnormalities and the reduced pollen fertility in an established species like *Derris indica* are important for understanding the mode of speciation in highly evolved plant species.

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1. Atchinson, E., *Amer. Jour. Bot.*, 1951, 36, 364.
2. Bennet, S. S. R., *JBNHS*, 1971, 68 (1), 302.
3. Patel, J. S. and Narayan, G. V., *Curr. Sci.*, 1937, 5, 479.
4. Sunder Raghavan, R. and Arora, C. M., *Proc. Indian Acad. Sci.*, 1958, 47 B, 352.
5. Sharma, A. K. and Sharma, A., *Chromosome Techniques* (Theory and Practice), 2nd Edition, Butterworth and Co., Ltd., London and University Park Press, Baltimore, 1972.
6. Tjio, J. M. and Levan, A., *Ann. Expt. Aula. Dei.*, 1950, 2 (1), 61.

A LEAF DISEASE OF POPLAR FROM KASHMIR

DURING the course of my investigations on the morphological variations of the conidia of *Spilocea pomi* Fr ex Fr. on different varieties of apples in the Kashmir valley, I came across a leaf spot disease on *Populus alba* in the Doabga orchard at Sopore. This fungus has been identified as *Cladosporium martianoffinum* Thüm., and has not been reported from India before. Some of the orchardists have mistaken this for the scab-causing organism.

Cladosporium martianoffianum Pilzfl. Sibir n. 474 *et* 653, Saccardo, *Syll. Fung. 4* : 357–358, 1886. Spots irregular, measuring 1–3 mm in diameter, hypophyllous, velutinous, deep olivaceous, often perforating the leaf ; stroma absent. Conidiophores macronematous, rarely branched, subfasciculate, deep brown, upto 45 μ long and 4–6 μ wide, conidiogenous cells polyblastic, indeterminate, terminal, pale brown to subhyaline ; conidia single, sometimes in short chains, broadly fusoid or obtuse, one septate, rarely single, pallid brown, 8–15 (–16) × 4 × 5 (–6) μ. Collected on living leaves of *Populus alba,* on 9th of July 1975, from Doabga apple orchard, Sopore, Baramula Dt., Kashmir (Herb. No. 195518 of the Commonwealth Mycological Institute, U.K.). This fungus was originally described on *Populus saveolens* and *P. laurifolia* from Siberia.

It may be of interest to mention here that *Fusicladium radiosum* (Lib.) Lind[1], was described on *Populus tremuloides* from Manitoba. This fungus was later on described as *Pollaccia radiosa* (Lib.) Bald. and Cliff, the perfect state being *Venturia macularis* (Fr.) E. Muller *et* Arx.

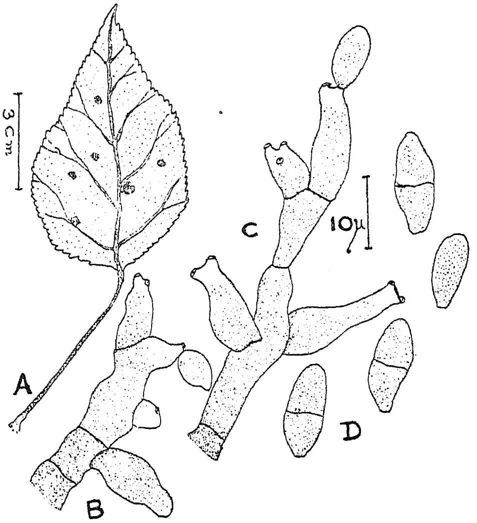

FIG. 1. A–D. *Cladosporium martianoffianum* Thüm. A. The undersurface of *Populus alba* leaf showing the fungal spots. B and C conidiophores and Conidia. D. Conidia.

The author is thankful to Mr. K. S. Ananthararaman, General Manager, F and P Division, Rallis India Ltd., for permission to publish this note and to Dr. M. B. Ellis of the Commonwealth Mycological Institute, Kew, Surrey, U.K., for confirming the identity of the fungus.

Research and Development V. AGNIHOTHRUDU.
Laboratories,
Fertilisers and Pesticides Division,
Rallis India Limited, Binfield,
87 Richmond Road,
Bangalore 560 025. *October* 3, 1975.

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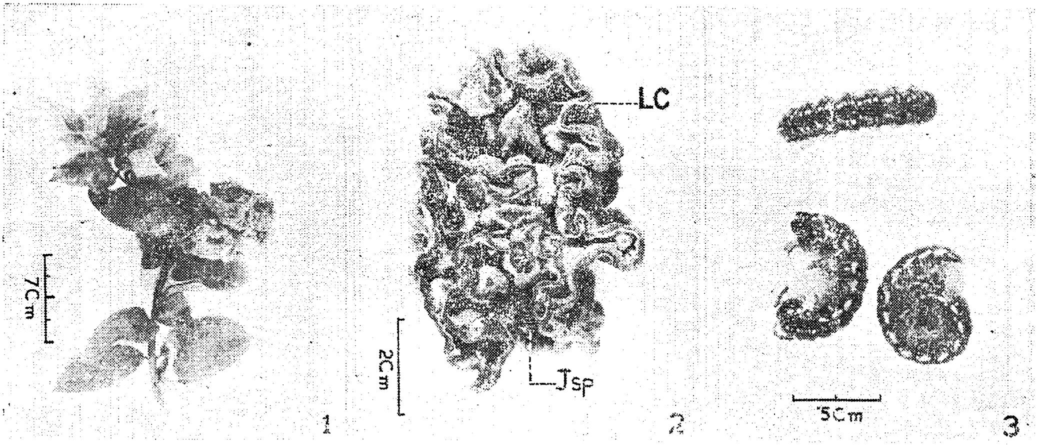
1. Sutton, B. C., *Mycological Papers*, C.M.I. 132, 1973, p. 91.

JONESPELTIS SPLENDIDUS (VERHOEFF) IN ASSOCIATION WITH *AUSTROTHRIPS COCHINCHINENSIS* KARNY IN THE GALLS OF *CALYCOPTERIS FLORIBUNDA* LAMK.

AN interesting association of the Diplopod, *Jonespeltis splendidus* (Verhoeff) with the gall-forming thrips, *Austrothrips cochinchinensis* Karny was observed during September 1975 in the Calicut University Campus. A survey of the literature revealed that no such association of *J. splendidus* with the gall-forming insects has so far been reported. Rao[7] and Ayyar[1] described the gall-forming thrips, *A. cochinchinensis* on *C. floribunda* as causing the formation of large, irregular, globose to elongate, convoluted, unilocular, hollow, greenish galls (Fig. 1).



FIGS. 1–3. Fig. 1. A branch of *C. floribunda* with mature gall showing the irregular, convoluted nature of the gall. Fig. 2. A gall cut open in the median longitudinal plane showing juveniles of *J. splendidus* in the labyrinthine interspaces of the larval cavity. (Jsp, juveniles of *J. splendidus*; LC, larval cavity.) Fig. 3. Juveniles of *J. splendidus* separated from the gall.

In the complex galleries of this gall, along with *A. cochinchinensis,* numerous other insects like *Androthrips ramakrishnae* Karny, predatory mites, and chalcidoid parasites *Megastigmus* sp., *Eurytoma* sp., have been reported earier[1,3,4,6]. Rao[7] also reported the caterpillar of an unidentified Pyralid moth as feeding on the gall tissue.

J. splendidus occurs normally in black moist soil, rich in decaying organic matter or in burrows in cowdung manure with adequate moisture. It feeds on the substratum in which it lives. While dissecting the mature galls for histopathological studies, the author found as many as eleven young ones of *J. splendidus* of varying sizes ranging from 0·7–2·1 cm in length and 1·0–2·0 mm in diameter in the tortuous labyrinthine interspaces of the interior of a mature gall (Figs. 2 and 3). No external opening indicating the point of entry of the larvae could be observed. Further, it is interesting to note that the larvae were found in a mature gall, formed on a branch 3 feet above the ground level.

The usual habitat of this millipede being the black moist soil rich in debris, it is concluded that the adult might have laid its eggs in the young bud of *C. floribunda* prior to the initiation of cecidogenesis so that the eggs got enclosed in the larval cavity during the development of this gall, and that these eggs might have hatched and developed into juveniles (Fig. 2). This view seems to be justified by the fact that the juveniles observed here are almost of the same growth period, possibly hatched from the eggs of a single adult that laid its eggs in the bud prior to gall initiation. No morphological abnormalities could be observed in the gall inhabited by the Diplopod.

A detailed examination of the gut contents of the larvae from the gall revealed green plant material under the microscope, indicating a change in their usual food preference. Millipedes are known to feed not only upon decaying organic matter, but also, under special circumstances, upon living plant tissue[2]. The juveniles of *J. splendidus*, as they are enclosed in the gall cavity which is isolated from the external environment, have no other way but to feed on the nutritive tissue lining the larval chamber. This supports the view of Baker[2] that the millipedes, under special circumstances, feed upon living plant tissue.

The author is thankful to Dr. B. K. Nayar, Professor and Head of the Department, to Dr. V. J. Philip, for his valuable guidance and encouragement and to Mr. C. Radhakrishnan, Department of Zoology, for helping me in examining the gut contents.

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1. Ayyar, T. V., *Mem. Dept. Agric. India* (*ent.*), 1928, 10, 239, 297.
2. Baker, A. N., In : *Myriapoda,* Ed., Blower, J. G., Acad. Press, London and N.Y., *Symp. Zool. Soc.,* London, 1974, 32, 621.
3. Mani, M. S., *J. Roy. Asiatic Soc. Bengal* (*Sci.*), 1948, **14**, 69.
4. —, "Cecidotheca Indica," *Agra. Univ. J. Res.* (*Sci.*), 1959, **8**, 91.
5. —, *Ecology of Plant Galls,* Dr. W. Junk, The Hague, 1964.
6. —, *Plant Galls of India,* Macmillan, India, 1973.
7. Rao, Y. R., *Agric. J. India,* 1924, **19**, 435.

SHORT SCIENTIFIC NOTES

Crystalline Components of *Clerodendron serratum*

Clerodendron serratum (L.) M. (Fam.: Verbenaceae) is a shrub with blue flowers, widely distributed in India. The root is used in medicine for fevers, rheumatism and dyspepsia and the leaves as external applications in cephalalgia and ophthalmia[1]. The isolation of D–mannitol from the root bark[2,3] and sapogenins from the bark[4] of this plant has been earlier reported. Subsequently, Rangaswami[5] raised a doubt about the botanical authenticity of this material, and hence the present investigation was taken up.

The roots obtained from Thanjavur (Tamil Nadu), yielded stigmasterol and a pigment, probably a quinone; the latter could not be characterised fully due to paucity of the material. Other constituents were non-crystalline.

The dry leaves of *C. serratum* (collected from Coimbatore) did not yield either stigmasterol or 24 S–ethyl cholesta–5, 22, 25-trien-3-β-ol[6]. Instead, it was found to contain α-spinasterol, m.p. 164–5° (acetate, m.p. 176–8°) identified by comparison with an authentic sample (m.m.p. and $AgNO_3$ impregnated co-TLC). The alcohol extract was found to contain (+)-catechin, luteolin and luteolin–7–0–β–D–glucuronide, all identified by direct comparison with authentic samples. We could not detect any mannitol in either of the materials studied.

We thank Mr. T. S. Srinivasan, Pharm Products, Thanjavur and Dr. J. Joseph, Botanical Survey of India, Coimbatore, for the genuine plant materials, Dr. S. K. Nigam, National Botanic Gardens, Lucknow, for authentic α-spinasterol and the Principal, JIPMER, for encouragement.

Dept. of Chemistry, A. G. Ramachandran Nair.
J.I.P.M.E.R., T. N. C. Vedantham.
Pondicherry 605 006, S. Sankara Subramanian.
December 12, 1975.

---

1. *Wealth of India, Raw Materials,* C.S.I.R., New Delhi, 1950, **2**, 232.
2. Garg, V. P. and Varma, S. C. L., *J. Pharm. Sci.,* 1967, **56**, 639.
3. Banerjee, S. K., Chakravarti, R. N., Sachdev, K. S. and Vasavada, S. A., *Phytochem.*, 1969, **8**, 515.
4. Rangaswami, S. and Sarangan, S., *Tetrahedron,* 1969, **25**, 3701.
5. Rangaswami, S. (Private communication).
6. Subramanian, S. S., Nair, A. G. R. and Vedantham, T. N. C., *Phytochem.*, 1973, **12**, 2078.

Transfer of Restorer Genes Over a New Genetic Background in Sunflower (*Helianthus annuus* L.)

Genetic male sterility in sunflower was first recorded in 1935 and in the subsequent years several reports have appeared. Recently, cytoplasmic male sterility was reported[2] in the back cross progenies of *Helianthus petiolaris* × *H. annuus*. The cytoplasmic male sterile line, 2 cm 183, its maintainer and the restorer line, BCZ 111 were obtained in 1973. The restorer line was very weak, stunted (about 30 cm) and possessed flower heads of about 6 cm diameter. Thus, it was not useful for the commercial hybrid seed production. Also, the hybrids derived from the cross, 2 cm 183 × BCZ 111 were in no way superior to the female parent. This cross was, however, helpful for the understanding of the genetic basis of fertility restoration. The segregation of F_2 generation, more closely approximated a 9:7 ratio, with two independent complementary dominant genes controlling restoration. Details are reported elsewhere.

The collections of sunflower cultures of Russian Canadian, Bulgarian and Romanian origin, available in this department, were tested for the restoration capacity, but none of them was found to possess the restorer genes. It has, therefore, become inevitable to transfer the restorer genes of BCZ 111 over a new genetic background. Since emasculation in sunflower is almost impossible, the male sterile hybrid of 2 cm 183 × E.C. 68415 was utilised as the female parent for the transfer of restorer genes from BCZ 111. The resultant hybrid of the triple crosss, (2 cm 183 × E.C. 68415) × BCZ 111, was male fertile. The second generation consisted of both male sterile and fertile plants. From the male fertile section, vigorous plants closely resembling E C. 68415 were selected and their fertility restoration capacity was tested in crosses with 2 cm 183. Majority of the tested plants have turned out to be either partial or complete restorers and some of them yielded vigorous hybrids. Thus, the transfer of restorer genes on to a new genetic background in sunflower has been achieved. Combining ability studies of crosses involving these derived restorers with 2 cm 183 male sterile line are in progress.

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1. Putt, E. D. and Heiser, B., Jr., *Crop Sci.*, 1966, **6**, 165.
2. Leclercq, P., *Ann. Amelior. Plantes*, 1969, **19**, 99 (*Bot. Abst.*, 1970, **51**, 10930).

Effect of Altozar, A Juvenile-Hormone Analogue, on *Coccinella septempunctata* Linnaeus

Altozar (ZR-512) is an effective growth regulator of the aphids. Recently, the authors found it very promising against the mustard aphid *Lipaphis erysimi* (Kaltenbach). Its safety to the final-instar grubs of *Coccinella septempunctata* Linnaeus, an important predator of this apid, was investigated. Thirty final-instar grubs were sprayed with 0·5 ml of 0·5% Altozar in acetone. These were fed on untreated mustard aphids. Another group of 15 untreated final-instar grubs was fed daily on aphids sprayed with Altozar at the said dose. Fifteen final-instar grubs which were sprayed with 0·5 ml acetone and offered untreated aphids served as control. In all the treatments the grubs pupated but amergence was only 13% in the first treatment and nil in the second treatment as against 67% in the control. No deformity was apparent in the grubs, pupae and adults, but Altozar definitely affected the development and caused mortality at the pupal stage. Bagley and Bauernfeind (1972) reported that *Chrysopa*, Hemipterous predators, Coccinellids, *Apanteles* and *Trichogramma* showed morphogenetic responses to the juvenile-hormone analogues, Roeller's and Bower's compounds.

We are thankful to M/s. Zoecon Corporation, Palo Alto. California, U.S.A., for supplying Altozar (ZR-512).

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Ludhiana, Punjab (India).
December 1, 1975.

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1. Bagley, R. W. and Bauernfeind, J. C., "Field experiences with juvenile hormone mimics". In *Insect Juvenile Hormones: Chemistry and Action*, Ed. Menn, J. J. and Beroza, M. Academic Press, New York and London, 1972, pp. 113–154.

A New Name for *Brachygrammatella indica* Khan (Hymenoptera: Trichogrammatidae)

Brachygrammalella aligarhensis Nom. n.

Brachygrammatella indica Khan, 1975, *Curr. Sci.*, **44**: 430. (Preoccupied by *Brachygrammatella indica* Viggiani and Hayat, 1974, *Boll. Lab. Ent. Agr. Portici* **31**: 150).

Viggiani and Hayat (1974) described the species *Brachygrammatella indica* which was reared from the eggs of *Oxyrachis* sp. at Manmad, Maharashtra. Later, Khan (1975) described another species under the same name *Brachygrammatella indica* which was reared from *Oxyrachis tarandus* Fabr. at Aligarh, Uttar Pradesh. Thus, *Brachygrammatella indica* Khan becomes a junior primary homonym of *Brachygrammatella indica* Viggiani and Hayat. Therefore, a new name *Brachygrammatella allgarhensis* is proposed for *Brachygrammatella indica* Khan, 1975 (nec. Viggiani and Hayat, 1974).

The author is indebted to Dr. S. Adam Shafee for supervising the work. He is also thankful to Dr. Mohammad Hayat for his useful suggestion.

Section of Entomology, M. YOUNUS KHAN.
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Aligarh Muslim University,
Aligarh, India, *December* 13, 1975.

Khan, M. Y., *Curr. Sci.*, 1975, **44**, 430.
Viggiani, G. and Hayat, M., *Boll. Lab. Ent. Agr. Portici*, 1974, **31**, 145.

Mycoflora Associated with Melon (*Cucumis melo* L.) Seeds

In the present work external and internal mycoflora of seeds of *Cucumis mejo* L. has been studied.

For the external mycoflora, the unsterilized seeds were plated on blotter paper and potato-dextrose-agar medium separately (ISTA, 1966) In order to detect the internal seed borne fungi, surface sterilized seeds were dissected and the testa, the cotyledons and the axis were plated separately on blotter paper and potato-dextrose-agar medium and incubated at 25° C

The 17 fungal isolates obtained and identified are:

Aspergillus flavus Link ex Fries, *A. niger* Van Tieghem, *A. terreus* Thom., *A. sulphureus* (Fres.) wehmer, *Cladosporium herbaram* (Pers.) Link. and *Penicillium* sp. were found in ecto and endophytic association.

Aspergillus flavns, *A. niger*, *A. terreus*, *A. tamarii* Kita, *A. versicolor* (Vuill.) Tiraboschi, *A. sulphureus*, *Alternaria tenuis* Nees., *Chaetomium globosum* Kunze & Schm., *Cladosporium herbarum* (Pers.) Link., *Penicillium* sp., *Curvularia* sp., *Helminthosporium* sp., *Fusarium* sp., *Monocillium* sp., *Molnilia* sp., *Cunninghamella* sp., and *Mycelia sterila* were found in endophytic association with seeds.

Aspergillus flavus, *A. terreus*, *Cladosporium herbarum* and *Penicillium* sp. were found associated with seed coat, cotyledons and axis of seeds.

Pathology Research Laboratory, M. N. GUPTA.
Department of Botany, V. K. AGRAWAL.
Agra College, Agra-282 002,
December 20, 1975.

---

1. Anonymous, "International rules and seed testing", *Proc. Int. Seed. Text. Assoc.*, 1966, **31**, 1.

Concerning Fungi and Actinomycetes of Rock Encrustations

Although abundant literature is available regarding isolation of fungi and actinomycetes from different habitats, there appears to be little data on the flora of these microorganisms from the dry barren mountainous rock encrustations. It is the purpose of this study to obtain such information.

During October 1973, samples of the barren rock encrustations from the Himalayan mountain at an altitude of 2833 m near Shillaroo (Himachal Pradesh) were collected with sterile precautions in presterilized polythene bags and brought to the laboratory for isolations. The soil-dilution-plate technique was employed. Fungi were isolated using Martin's medium (Martin, 1950), and acticnmycetes with starch-casein medium (Kuster and Williams, 1964). Incubations were made for 7 days at 24 ± 1° C for fungi and 29 ± 1° C for actinomycetes. The discrete colonies from the plates were transferred on fresh media and pure cultures were obtained.

The species of fungi included *Aspergillus niveus* Blochwitz, *A. terreus* Thom, *A. ustus* (Bainier) Thom and Church, *A. versicolor* (Vuillemin) Tirab., *Botryotrichum piluliferum* Sacc. and March., *Chaetomium globosum* Kunze, *Claadosporium* sp., *Karnia* sp., *Papulospora* sp., *Penicillium citrinum* Thom, *P. variabile* Sopp, *Scobllcobacidlum constrictum* Abbott and *Sepedonium* sp.

The species of actinomycetes found were, *viz.*, *Streptomyces* sp., *S. griseoflavus* (Krainsky) Waksman and Henrici, *S. griseus* Waksman and Henrici, *S. olivaceous* (Waksman) Waksman and Henrici and *S. ramulosus* Ettlinger, L., Gaumann, E., Huter, R., Neipp, L., Prelog, V. and Zahner, H.

The presence of these micro-organisms under such unfavourable growth conditions is noteworthy.

The author is thankful to the Director, Commonwealth Mycological Institute, Kew, for identification of the cultures. The research facility received at Central Potato Research Institute, Simla, is sincerely acknowledged.

Central Potato Research Institute, R. P. RAI.*
Simla-1 (H.P.), *July* 9, 1975.

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* Present address: Dr. R. P. Rai, National Environmental Engineering Research Institute, Nagpur-20.

1. Kuster, E. and Williams, S. T., *Nature, Lond.*, 1964, **202**, 928.
2. Martin, J. P., *Soil. Sci.*, 1950, **69**, 215.

Foot and Bulb Rot of *Ornithogalum virens*

During the September–November period of 1973 and 1974, a severe foot and bulb rot of *Ornithogalum virens* L., an ornamental plant, was observed on the Campus of S.V. University, Tirupati (A.P.). The disease incidence was high at the seedling stage. With age, the plants appeared to be less susceptible.

To begin with, the water-soaked spots, (2–3 mm dia) were observed at the foot of the infected plants. Subsequently, the spots enlarged coalesced and turned necrotic, encircling the foot region as a result of which the aerial part of the plant collapsed. This was followed by the complete rotting of the bulb within 10–12 days. At this stage clusters of dark brown sclerotia (1–2 mm in dia) were seen on the surface of the rotting bulb.

The causal organism was isolated from the diseased tissue and its pathogenicity established. Reisolations from the inoculated plants yielded the fungus similar to the parent culture. The fungus was identified as *Sclerotium rolfsii* Sacc.

Host range studies were conducted employing one month old seedlings of *Allium cepa* L., *A. sativum* L., *Gloriosa superba* L., *Chlorophytum* sp., *Sanseviera roxburghiana* Thunb. *Scilla indica* L. and *Asparagus racemosus* L., all of which belong to the same family

Lilliaceae. The disease symptoms were observed only in *Chlorophytum* sp., *Sansveria roxburghiana* and *Scilla indica* and within 3–5 days after inoculation. The foot and bulb rot of *Ornithogalum virens* incited by *Sclerotium rolfsii* is a new record for India.

The authors are indebted to Dr. Y. R. Sarma for help.

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Tirupati, A.P., *July* 25, 1975.

Occurrence of *Ephelis oryzae* Syd. on Pearl Millet

During the *Kharif* season of 1975, a crop of pearl millet, *Pennisetum typhoides* (Burm.) Stapf. and Hubb. variety HB-3, was grown besides a fox tail millet, *Setaria italica* Beauv., the crop was affected with *Ephelis oryzae* Syd. A single earhead of pearl millet bordering fox tail millet crop showed symptoms similar to that of *Ephelis* infection. The florets in the lower half of the affected head were grayish and found glued and pressed towards the rachis. Microscopic examination of the fungus revealed the spores to be acicular hyaline and measured $15 \cdot 3–24 \cdot 3 \times 1 \cdot 4–1 \cdot 8\,\mu$ with an average length of $19 \cdot 26\mu$ which were very close as to the spore size of *E. oryzae* on *Setaria italica* as reported by Venkatakrishnaiah (1952). Govindu and Thirumalachar (1961) have recorded *Ephelis* on *Pennisetum hohenackeri* Hocst and *P. alopecuras* Steud. has been reported as a host of *E. oryzae* by Misra and Pall (1975). There is no report of *Ephelis* sp. on *Pennisetum typhoides*. Based on the spore dimensions, *P. typhoides* is considered as a new host of *E. oryzae* from India.

The specimen is deposited in the mycological herbarium of the Department of Plant Pathology, University of Agricultural Sciences, Bangalore (Mysp # 1958).

The authors wish to thank the authorities of the University of Agricultural Sciences, Bangalore, and the Indian Council of Agricultural. Research, for facilities and financial assistance repectively.

Department of Plant H. R. Reddy
Patholgoy, K. A. Lucy Channamma.
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December 18, 1975.

---

1. Govindu, H. C. and Thirumalachar, M. J., *Mycopath. et Mycol. appl.*, 1961, **14**, 189.
2. Misra, R. P. and Pall, B. S., *Curr. Sci.*, 1975, **44** (23), 869.
3. Venkatakrishnaiah, N. S., *Phytopathology*, 1952, **42**, 634.

Leaf Spot Disease of *Rauwolfia serpentina* Caused by *Corynespora cassiicola* (Berk. & Curt.) Wei from Manipur

A severe leaf spot disease was observed during August 1975 on the older leaves of a potted plant of *Rauwolfia serpentina* (Linn.) Benth. ex. Kurz. (Apocynaceae) in the Botany Garden, D.M. College, Imphal, Central Manipur District, Manipur State. The disease manifests itself in the form of deep dark brown spots with distinct concentric rings, 1–16 mm. in diameter. Infection starts from the margin rarely at the midrib, more at the apical portion, lower surface faint dark tan with concentric Zones surrounded by brownish yellow margin. Number of spots per leaf varies (11–18). occasionally spots enlarged and coalescenced forming bigger patches.

The pathogen was isolated in pure form from the infected spots on Czapek's agar and identified[1-4] as *Corynespora cassiicola* (Berk. and Curt.) Wei. Pathogenicity of the fungus was successfully proved by spraying the spore suspension on the healthy injured leaves of the host. Uninjured leaves inoculated with the fungus did not produce spots.

Available literature[5-7] shows that the fungus was not obtained earlier from *R. serpentina* and hence this is the first record of the fungus for Manipur. Two of the infected leaves have been deposited at C.M.I., Kew, Surrey, England, as IMI. No. 196571.

The senior author expresses his sincere thanks to Rev. Br. R. B. Vieyra, Principal and Dr. K. S. Singh, Head, Botany Department, St. Edmund's College, Shillong for laboratory facilities and encouragement respectively. We are also thankful to Dr. M. B. Ellis, C.M.I., Ferrylane, Kew, England, for confirming identity of the fungus.

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Gauhati 781 014, *December* 10, 1975.

---

1. Ainsworth, G. C. and Bisby, G. R., *Dictionary of the Fungi*, C.M.I., Kew, Surrey, 1971, p. 136.
2. Barnett, H. L. and Hunter, B. B., *Illustrated Genera of Imperfect Fungi*, Burgess Pub. Co., Minnesota, U.S.A., 1972, p. 116.
3. Subramanian, C. V., *Hyphomycetes*, I.C.A.R., New Delhi, 1971, p. 735.
4. Wei, C. T., *Mycological Papers*, C.M.I., Kew, 1950, **34**, 5.
5. Dubey, G. S., *Curr. Sci.*, 1975, **44** (22), 825.
6. Mukerji, K. G. and Juneja, R. C., *Fungi of India*, Emkay Pub., Delhi, 1975.
7. Subrahmanyam, P. and Sarma, Y. R., *Curr. Sci.*, 1974, **43** (13), 429.

REVIEWS AND NOTICES OF BOOKS

Interfacial Phenomena in Metals and Alloys. By Lawrence E. Murr. (Addison-Wesley Publishing Co., Massachusetts, U.S.A.), 1975. Pp. xiv + 376. Price $ 24.50.

It is well known that interfaces in solids determine many of the properties and behaviour of materials. Interfaces are generally regarded as two-dimensional imperfections, but are more difficult to study, than the role of dislocations and point defects. External and internal interfaces are intimately involved in oxidation and corrosion, friction and wear, plastic flow and fracture, intergranular embrittlement, phase transformations, nucleation and growth processes, catalysis and heterogeneous reactions, recrystallization and growth, adhesion and sintering, etc., their importance in fine-particle processing, polymer technology, biocompatibility and virtually all types of materials processing is now being appreciated by all. And many new tools are available for their studies, such as FIM, Auger spectroscopy, LEED, electron microscopy, thin film techniques, and photoelectric and semiconductor effects. It is then a great boon to materials scientists that a book is now available on this important topic. I must say, the book is written in an excellent style of an advanced textbook, which makes it highly useful to both beginners as well as experts. The bias is of course on metals and alloys, and in this respect, the reviewer is slightly disappointed.

In the first chapter, the Thermodynamics of Solid Interfaces are introduced with a large number of supporting references. The second chapter deals with Interfacial Equilibria and the third chapter is on Interfacial Free Energy (78 pages, with 191 references, 39 figures, 10 tables); the author has been very successful in presenting the latter chapter; but the experimental details seem to be inadequate. The Structure of Interfaces is presented in Chapter 4, with 222 references, 66 figures, and 7 tables. The last but the most important chapter on Properties of Interfaces is very lucidly written and substantiated with 264 references, and 58 figures. Each chapter ends with a set of problems.

It can certainly be said that the book is written in the style of a textbook, and to a certain extent, it meets the requirements of a reference volume. A perusal of the book will give an immense insight into a variety of metallugical phenomena, and it is highly recommended for all libraries.

K. I. VASU.

Ultra-violet and Visible Spectroscopy, Chemical Applications (Third Edition). By C. N. R. Rao. (Butterworths, London), 1975. Pp. xiii + 242. Price £ 9.00.

This is a revised edition of the book first published in 1967. New information has been included in a number of chapters, *e.g.*, on Photoelectron spectra (Chapter 9), on Circular dichroism and liquid crystals (Chapter 12). In its thirteen chapters, the book deals with a large number of topics and phenomena, concerning ultraviolet and visible spectroscopy. Because of the short compass, it has not been possible for the author to deal with some more important topies in sufficient detail and depth.

A few minor errors given below have been noticed: On page 34, last but one line, Table 3·8, may be changed to Table 3.9. On page 102, the description of tautomerism is rather too restrictive. On page 126, line 16 the sentence would read better without 'recently'. On page 105, the finding 'that in a keto-enol tautomeric equilibrium, enol content is decreased in polar and hydrogen bonding solvents' should be credited to K. H. Meyer, *Ber* 45, 2843 (1912).

The popularity of this book can be gauged by the fact that it has been translated into Russian, Spanish and Japanese languages.

The book serves as a good general introduction and guide to persons interested in the applications of ultra-violet and visible spectroscopy.

M. V. BHATT.

The Spectroscopy of Flames (Second Edition). By A. G. Gaydon. (Chapman and Hall Ltd., 11 New Fetter Lane, London, EC4P 4EE), 1975. Pp. xii + 412. Price: £ 8.00.

Spectroscopy is one of the most important tools to investigate combustion processes in flames. The evolution of an entirely new branch of flame spectroscopy is due to the new developments in the field of spectro-chemical analysis. Recently developed atomic absorption and atomic fluorescence spectroscopy are epoch-making advances in spectrochemical analysis. At this juncture second edition of A. G. Gaydon's *The Spectroscopy of Flames* will acquire a warm welcome amongst research worker, is this field. The book is a thorough revision of the first edition published in 1957. Professor Gaydon has been working in the field of flame spectroscopy for about two scores of years and is one of the authorities to write on the subject.

The book deals with the use of spectroscopic observations to interpret combustion processes in flames asd flame structures. A brief study made on explosion flames and engines is given in a separate chapter. It serves a useful purpose to those working in industry, especially when the author takes subjects like diesel engines, exhaust flames, furnance flames and solid propellants. Studies of hydrogen flame and carbon monoxide flame are remarkable. Comments on the use of flames for spectro-chemical analysis including atomic absorption and atomic fluorescence make the book upto date. Inclusion of a detailed study of flash photolysis and chemiluminescence might have enhanced the scope of the book.

The structure of the book is such that it is useful to research workers as well as to beginners and specialists. A brief theory of molecular spectra is also presented without any mathematical treatment which makes the book self-contained, when the understanding of the subject is concerned.

An extensive bibliography and an appendix giving tables of wavelengths of all main band spectra frequently found in flames, and plates of spectrograms make the book valuable for reference purposes.

M. M. PATEL.

Physics of Quantum Electronics (Vol. 2.): *Laser Applications to Optics and Spectroscopy* (Edited by S. F. Jacobs, M. Sargent III, J. F. Scott and M. O. Scully. (Published by Addison-Wesley Publishing Company, Inc., Advanced Book Program, Reading, Massachusetts), 1975. Pp. xii + 510. Price $ 23.50.

This is the second volume of the serial publication *Physics of Quantum Electronics*. It is based on lectures delivered by specialists at the Physics of Quantum Electronics summer school at the Crystal Mountain (Washington) session in 1973. The topics covered are tunable diode lasers and their applications, Dye lasers, Spin-Flip light scattering and spin flip lasers, harmonic generation in gases and production of ultraviolet and vacuum ultraviolet radiation, Integrated optics, Nonlinear effects in optical fibers, application of photon statistics and photon correlation, light scattering, made-locked lasers and ultrashort light pulses, stabilised lasers and applications, adiabatic following and nonlinear spectroscopy. Diode lasers on account of their tunability, narrow, spectral line width, high degree of stability, low input power requirements and structural simplicity are finding wide application in high resolution gas spectroscopy and photochemistry. The first paper summarizes the present state of the art with regard to lead salt diodes, Ga A_s–Al_x Ga_{1-x} A_s heterostructure lasers. Although the review is brief, adequate references have been given to cover the entire subject. As tunable laser sources are the basis of much of the recent developments in spectroscopy, the subsequent lectures are devoted to dye lasers and spin flip lasers. The chapter on dye lasers is quite comprehensive and besides dealing with the principles of dye lasers, attention has been drawn to scope for further research in the physical chemistry of organic dyes and their potentiality towards applications in biology, medicine and communications. The chapter in spin flip lasers and spin flip scattering covers the basic theory and gives several examples from the precision spectroscopy that has become possible and emphasizes its advantages over EPR in studying even heavily doped semiconductors. In Chapter 4 the nonlinear optical techniques that make use of phase matched mixtures of metal vapours and inert gases have been described. At the end, the interesting application of holographic technique at soft X-ray wavelength and its future scope have been dealt with.

Integrated optics is a subject of recent origin and its growth had to wait for the development of thin and thick films and fibre optics. However during the past ten years considerable progress has taken place in research on monolithic integrated optics and nonlinear effects in optical fibres even at low power levels and fiber communication. The present state of the art in these fields have been covered. The development of highly stable and intense lasers, obviously, had their maximum impact on spectroscopy and scattering. It is therefore natural that a large portion of the lectures is devoted to topics like nonlinear spectroscopy, Raman, Brillonin and Rayleigh scattering and attention has been drawn to the areas where further scope for application exists, especially in biomolecular topics. The utility of the present Vol. 2, is considerably enhanced by the inclusion of the reprints on mode-locked lasers and ultrasonic light pulses, stabilized lasers and their applications.

P. S. N.

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CHARACTERISATION OF PROTEIN-CHAIN-FOLDING USING PEPTIDE PLANE NORMALS

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THE general problem of representation of the structural data from X-ray analysis of protein crystals has recently been considered in this laboratory and some new methods have been proposed[1–2]. Broadly two types of representations have been proposed, namely, one-parameter and two-parameter representations which are aimed at extracting certain specific types of information about the chain folding. The basis of the new parameters itself is on the consideration of a set of four consecutive C^α atoms, $C^\alpha{}_{i-1}$, $C_i{}^\alpha$, $C^\alpha{}_{i+1}$, $C^\alpha{}_{i+2}$ and defining angles involving the virtual bonds connecting the consecutive C^α atoms (such as the virtual bond-angle $C^\alpha{}_{i-1} - \hat{C}_i{}^\alpha — C^\alpha{}_{i+1} = \delta$ and the virtual-bond dehedral angle θ around the virtual bond $C_i{}^\alpha — C^\alpha{}_{i+1}$) and also defining a local helical axis of the segment $C^\alpha{}_{i-1} — C_i{}^\alpha — C^\alpha{}_{i+1} — C^\alpha{}_{j+\cdot 2}$. It has been demonstrated that the use of these new parameters in different types of one-parameter representations (θ and δ-chain plots) and two-parameter representations such as η_{ij} plot, as well the two-dimensional representation such as stereographic projection lead to useful information on chain folding characteristics. It may be noted that in contrast to the above, the conventional two-parameter representation, namely, the (ϕ, ψ) diagram[3] deals with torsion angles around real bonds, namely N—C^α and C^α—C of a pair of peptide planes linked at a given C^α atom. The basis of the (ϕ, ψ) representation is the simple fact that the peptide units are planar* and any two successive planes take their relative orientation by virtue of the freedom of rotation along the single bonds involving the linking C^α atom. It would appear that the physical reality that the peptide unit, for all practical purposes, is planar and rigid could be exploited further to devise parameters which would lead to more such representations. Thus, one may define vectors which are normal to these planes and use these for the purposes of representations. The object of this communication is to point out that these vectors appropriately defined enable us to carry out many of the types of analyses discussed earlier. For instance the chain plots can be obtained using ν_i which is the angle between successive plane normals, say, $\boldsymbol{n}_{i-1}$ and $\boldsymbol{n}_i$**. So also a two-parameter representation, namely, ν_{ij} similar to η_{ij} is possible. Lastly the direction n_i can be used for a stereographic projection.

Chain-folding Characterisation Using Peptide-plane Normals:

Consider the polypeptide chain as in Fig. 1. For all practical purposes it is assumed that all the backbone atom $C_i{}^\alpha$, C_i, O_i, N_{i+1}, H_{i+1} lie in a plane, whose unit normal is denoted by n_i. It is readily obtained by taking the cross-product of the vectors $\overrightarrow{C_i{}^\alpha - C_i}$ and $\overrightarrow{C_i - N_{i+1}}$ (Fig. 1) and normalising the resultant vector. The dot product of two such unit vector $\boldsymbol{n}_{i-1}$ and $\boldsymbol{n}_i$ leads to the angle between the pair of peptide planes. Following our earlier conventions[2] this may be denoted by ν_i. It is interesting to note that this angle, being the angle between two planes, can be considered as a dehedral angle and hence in principle it should be possible to have the full range of − 180° to +180°. This is readily visualised by the fact that for any relative orientation of the two planes one can imagine an axis (which is actually along the direction of the line of intersection of the two planes) about which the two planes may be assumed to be rotated by the torsion angle. We may thus proceed to assign a sign to the angle ν by a suitable method as is done normally for torsion angles. Thus let $\boldsymbol{n}_{i-1} \times \boldsymbol{n}_i = \boldsymbol{r}_i$, so that $\boldsymbol{r}_i$ may be seen to lie along the line of intersection of the two planes. The vectors s_i defined by $\overrightarrow{C^\alpha{}_{i-1} — C^\alpha{}_{i+1}}$ may be taken as a reference direction. Thus for assigning a sign we may stipulate a rule based on comparison of the vector $\boldsymbol{r}_i$ and s_i. In the special case of $\boldsymbol{r}_i$ and $\boldsymbol{s}_i$ being collinear, one may take ν_i to be positive if $\boldsymbol{r}_i$ and s_i are parallel and negative if they are antiparallel. However since $\boldsymbol{r}_i$ and $\boldsymbol{s}_i$ need not always be collinear, one may find the angle between the vector $\boldsymbol{r}_i$ and s_i by taking dot product $s_i — s_i = \mu_i$.

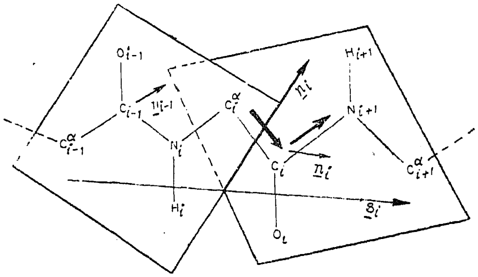

FIG. 1. Schematic diagram of a peptide chain showing vectors used for the characterisation of chain folding (*see* text for details).

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* In actual cases the peptide units are slightly non-planar [*see*, for example, references (4) and (5)].

** Sasisekharan[6] has considered the angle between two peptide planes in a different context but has taken only the acute angle. It may be noted that, in principle, since we are dealing with the angle between two planes a full range of −180° to +180° can be realised which preserves the sense of folding similar to the angle parameter θ considered earlier[1,2].

The sign for ν_i is chosen positive or negative depending on the angle μ_i being acute or obtuse, respectively.

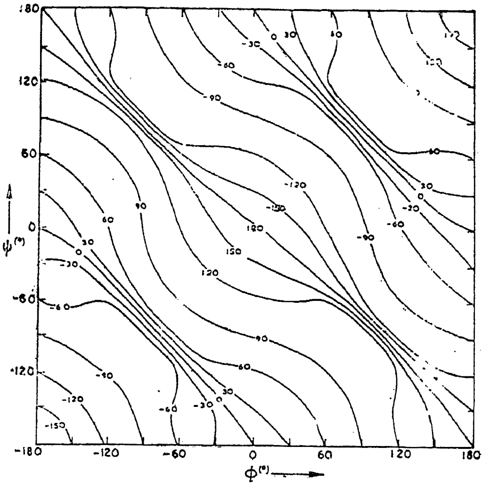

FIG. 2

FIG. 2. Equi ν_i-contours (*see* text for definition) in the (ϕ, ψ) plane.

Physically it may be seen that the above method of assigning the sign is equivalent to projecting the point C^{α}_{i-1} and C^{α}_{i+1} on to the line of intersection of the two planes as given by the vector r_i, and comparing the projected vector of $\overrightarrow{C^{\alpha}_{i-1}—C^{\alpha}_{i+1}}$ with r_i. While it might be possible to adopt for the above comparison any vector other than $\overrightarrow{C^{\alpha}_{i-1}—C^{\alpha}_{i+1}}$, we have chosen this here, since, as far as backbone atoms are concerned C^{α} atoms act as convenient reference points and also the progress of the chain is readily reckoned by tracing their path.

Figure 2 gives the equi-ν contours in the (ϕ, ψ) plane. The 'centrosymmetric' pattern of the contours is also readily understood. For example, the conformation $\phi = -120°$, $\psi = -60°$ and $\phi = +120°$, $\psi = +60°$ have ν values of $-60°$ and $+60°$ respectively.

A more detailed application of these ideas along the lines of our earlier analysis, including statisticaa. distribution of the angle ν_i is in progress and will be reported in due course.

1. Srinivasan, R and Balasubramanian, R., *Curr. Sci.*, 1975, **44**, 649.
2. —, — and Rajan, S. S., *J. Mol. Biol.*, 1975, **98**, 739.
3. Ramachandran, G. N. and Sasisekharan, V., *Adv. Protein Chem.*, 1968, **23**, 323.
4. Balasubramanian, R. and Ramakrishnan, C., *Int. J. Peptide Protein Res.*, 1972, **4**, 91.
5. Ramachandran, G. N. and Kolaskar, A. S., *Biochim. Biophys. Acta*, 1973, **303**, 385.
6. Sasisekharan, V., In: *The Jerusalem Symposia on Quantum Chemistry and Biochemistry* (Bergmann, E. D. and Pullman, B., Eds.), Jerusalem Academic Press, 1973, **5**, 36.

FIVE COORDINATE OXOVANADIUM (IV) COMPLEXES

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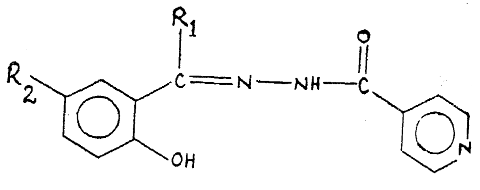
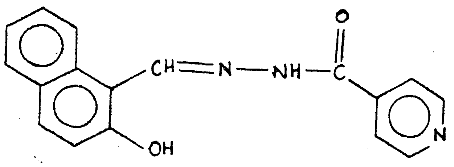
INTRODUCTION

It is well known that the hydrazide residue in the hydrazones can react with metal ion either in keto or in the enol form[1]. Hydrazones derived from isonicotinoyl hydrazide have been used as chelating agents[2]. Complexes of oxovanadium (IV) and oxovanadium (V) with benzimidazoles[3] and Schiff bases[4] have been studied. It is apparent from the literature that there are quite a few scattered examples of vanadium complexes with hydrazones[5]. It is therefore appropriate to study complex formation of oxovanadium (IV) with the following hydrazones.





	R_1	R_2		R_1	R_2
I	H	H	V	CH_3	H
II	H	Cl	VI	CH_3	Cl
III	H	CH_3	VII	CH_3	CH_3

EXPERIMENTAL

Vanadyl sulphate, isonicotinic acid hydrazide were of reagent grade. The substituted salicylaldehydes and acetophenones were prepared by standard methods[6,7]. Hydrazones were prepared by mixing equimolar mixture of hydrazide and aldehyde or ketone in the absolute ethanol. Tht solid products obtained were recrystallised from ethanol.

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* For correspondence.

The complexes were prepared by mixing the alcoholic solution of vanadyl sulphate (0·1 mole) with alcoholic solution of ligand (0·2 mole). The mixture was heated on a water bath for an hour. The precipitated complex was filtered and washed successively with small portions of warm aqueous alcohol to remove the excess of the ligand. The complexes are then dried in a desiccator over phosphorus pentoxide.

Vanadium in the complexes was estimated as V_2O_5, by precipitating vanadium hydroxide and igniting it to pentoxide. Nitrogen was estimated by the microanalytical method.

The elemental analysis is shown in Table I.

Physical Measurements.—The conductance measurements in DMF were done on Philips PR 9500 conductivity bridge (cell constant 0·829 cm^{-1}).

The magnetic susceptibilities of the complexes at room temperature were determined on a Gouy balance using mercury cobalt thiocyanate as calibrant.

The infrared spectra of the ligands and the complexes in nujol mull were recorded on a Perkin–Elmer 337 spectrophotometer in the region 4000–650 cm^{-1}.

RESULTS AND DISCUSSION

The complexes are dark brown in colour and are amorphous in nature. These are sparingly soluble in DMSO, DMF and pyridine. However the satisfactory electronic spectra were not obtained with solution. Limited solubility of these complexes in common organic solvents did not permit the determination of molecular weights.

The results of elemental analysis shown in Table I agree well with 1 : 2 stoichiometry. Oxovanadium (IV) sulphate loses its sulphate ion during the course of reaction with hydrazones. The proton of the *o*-hydroxy group is replaced by vanadium. The solution of these complexes in DMF do not show measurable conductance, hence the complexes are non-electrolytes.

TABLE I

Ligand No.	Complex No.	Empirical formula	Metal %	Nitrogen %	Magnetic moment at 298° K μeff B.M.	Important IR frequencies (cm.$^{-1}$)
I	VIII	$VO\,(C_{13}H_{10}O_2N_3)_2$	9·61 (9·31)	15·86 (15·35)	1·68	1675, 1605, 1285, 895
II	IX	$VO\,(C_{13}H_9O_2N_3Cl)_2$	8·21 (8·26)	13·32 (13·61)	1·70	1675, 1605, 1285, 890
III	X	$VO\,(C_{14}H_{12}O_2N_3)_2$	8·68 (8·86)	14·00 (14·55)	1·82	1670, 1612, 1275, 980
IV	XI	$VO\,(C_{17}H_{12}O_2N_3)_2$	8·21 (7·88)	12·30 (12·99)	1·77	1670, 1612, 1325, 950
V	XII	$VO\,(C_{14}H_{12}O_2N_3)_2$	8·90 (8·86)	14·20 (14·55)	1·79	1650, 1595, 1300, 960
VI	XIII	$VO\,(C_{14}H_{11}O_2N_3Cl)_2$	8·02 (7·92)	12·82 (13·06)	1·84	1650, 1610, 1295, 960
VII	XIV	$VO\,(C_{15}H_{14}O_2N_3)_2$	8·38 (8·48)	13·80 (13·89)	1·80	1640, 1612, 1290, 960

Values in parentheses are calculated ones.

Magnetic Properties.—The magnetic moments (Table I) corrected for the ligands determined for these complexes are in the range 1·68 to 1·84 B.M. This value is in close agreement with the values reported[8] for oxovanadium (IV) containing one unpaired electron. It can be concluded that vanadium (IV) has not been oxidised to vanadium (V) state during the course of reaction.

Infrared Spectra.—The IR spectra are interpreted based on the previous assignments[1,9,10,11]. In the spectra of the ligands I–IV and V–VII medium intensity band observed in the region 3200–3170 cm^{-1} and 3130–3100 cm^{-1} respectively is assigned to NH stretching. The band observed in the region 2800–2700 cm^{-1} in ligands I-IV and 3480–3460 cm^{-1} in ligands V–VII is assigned to intramolecular hydrogen bonded OH. In the complexes the NH band is retained but OH band disappears, indicating the replacement of H of hydroxy group by metal.

The strong band around 1675–1650 cm^{-1} ($\nu C = O$) is present in ligands and is retained in the complexes. Three to four bands of medium to strong intensity are observed in the region 1625–1500 cm^{-1}. These arise from various modes of vibration such as $\nu C{=}N$, $\nu C{=}C$, δOH, $\delta N{-}H$, etc., and their conclusive assignment is rather difficult. The band in the region 1625–1610 cm^{-1} is strong and shows a measurable shift to lower frequency by about 15–25 cm^{-1} in the complexes. Taking into consideration its intensity, the donor capacity of nitrogen and previous assignments[1,9-11]. This may reasonably be assigned to $\nu C{=}N$ and the shift may be due to the coordination of nitrogen to the metal. In the complexes IX, XI–XIV, the strong band observed around 960 cm^{-1} and in the complexes VIII and X the band around 890 cm^{-1} is assigned to $\nu V{=}O$ this probably suggests the monomeric nature of oxovanadium species[12,13].

On the basis of above observations it may be concluded that ligands have reacted in keto form and vanadium has coordination number five in the above complexes. Generally five coordinate oxovanadium (IV) complexes have square pyramidal structures.

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1. Narang, K. K. and Miss Aggarwal and references therein, *Inorganic Chim. Acta*, 1974, **9**, 137.
2. Amal, H. and Ergenc., *C.A.*, 1963, **59**, 3515 *d*; *C.A.*, 1964, **60**, 80025 *a*; Doadric, A. and Garcia, Cargo, *C.A.*, 1966, **65**, 16474 *a*.
3. Biradar, N. S., Devale, S. and Kulkarni, V. H., *Curr. Sci.*, 1973, **42**, 125.
4. — and Kulkarni, V. H., *Rev., Roum. Chim.*, 1971, **16**, 1203; 1972, **17**, 481; 1973, **18**, 63.
5. Farben tabriken Bayer, A. G., *C.A.*, 1957, **51**, 18006 *b*.
6. Duff, J. C., *J. Chem. Soc.*, 1941, p. 547.
7. —, *Organic Synthesis Collectives*, 1963, **4**, 106.
8. Dey, K., Chatterjee, K. K., and Sen, S. K., *J. Indian Chem. Soc.*, 1973, **50**, 167.
9. Rao, C. N. R., *Chemical Applications of Infrared Spectroscopy*, Academic Press, New York, London, 1962.
10. Clougherty, L. E., Sousa, J. H. and Wyman, G. M., *J. Org. Chem.*, 1957, **22**, 462.
11. Kovacic, J. E., *Spectrochim. Acta*, 1967, **23A**, 183.
12. Barraclough, C. G., Lewis, J. and Nyholm, R. S., *J. Chem. Soc.*, 1959, p. 3552.
13. Selbin, J., *J. Chem. Educ.*, 1964, **41**, 86.

ANALYTICAL REACTIONS OF BARBITURIC ACIDS

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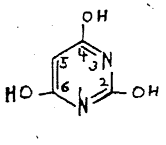
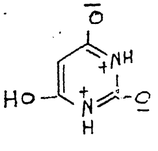
ABSTRACT

Analytical reactions of barbituric acids used in their identification and also the role of the acids as analytical reagents have been reviewed.

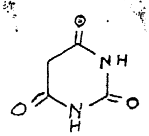
THE name barbituric acid was given to the compound by Alfred von Baeyer who possibly discovered it on St. Barbara's day or wanted to honour his dear friend. It exists in the tautomeric forms, I–III, though the X-ray studies[1] of the solid favour the structure (III). Its preparation involves the condensation of malonic acid with urea in the presence of $POCl_3$. It melts at 248–252° C (*d*). Though the acid itself is not biologically active, substituents especially at 5–C atom make it so. Depressant activity of the compounds increases with heavier substituents. Also the compounds become rapidly more toxic when the molecular weights go beyond 250–300. The most commonly used barbiturates for medicinal purposes contain substituents with 6–10 C–atoms. In a recent article[2] (incidently one of the authors is Barbara) the biochemical and physiological aspects of barbiturates have been summarized. It is now proposed to review the subject from analytical standpoint.



(I)



(II)



(III)

Barbituric, thiobarbituric, diphenylbarbituric and diphenylthiobarbituric acids have been used[3-5] to quantitatively precipitate and estimate furfural. The method is superior to that using phloroglucinol. The insoluble precipitate remains unaffected in dilute mineral acids. Large excess of the reagent should be avoided to eliminate its adsorption on the precipitate.

Pyridines have been indirectly determined[6-8] with barbituric acid. The method is based on the opening of pyridine ring by CNCl to form glutaconic dialdehyde which condenses with barbituric acid to give coloured polymethine dye suitable for photometric determination. Absorption maxima for the dyes corresponding to pyridine, picolines (α-, β-, and γ-), isonicotinic acid, isonicotinoyl glycine and isonicotamide lie at 578, 600–615, 600, 600–615 and 615 nm respectively. The method is extended[9] to determine pyridine and nicotine simultaneously. Chlorine[10], cyanide and thiocyanate[11] and CNS^- in cyanamide derivatives[12] have also been determined as they can liberate CNCl which subsequently reacts with pyridine and barbituric acid to form the dye. Traces of chlorine down to 2 μg are determined[13] in layers of selenium rectifier cells using this technique. Nikethamide has been colorimetrically determined[14] by measuring the absorbance of the pink solution formed when it reacts with CNBr and barbituric acid.

Copper (II) and cobalt (II) form coloured complexes with some barbituric acids[15] (Table I). Limit of identification for cobalt complexation with thiobarbituric acid is 10 μg. The reaction is used[16] for detection of barbiturates. Sodium ethyl-1-methylbutylthiobarbiturate reacts with ammonical Cu (II) solution forming emerald green coloured complex[17]. The reaction though specific is not much of analytical interest beacuse of instability of the complex. Ethyl-5-(1-methylbutyl) thiobarbituric acid is, however, identified[18] using its complexation with copper (limit of identification : 40 μg ; limit of dilution : 1 : 20,000). Silver nitrate reacts with barbituric acid and its 5, 5-diethyl derivative giving yellow precipitate soluble in alkali (λ_{max} 390–400 nm)[19]. The reaction is used for volumetric determination[20,21] and chromatographic identification[22] of barbituric acids. Equimolar complexes formed between UO_2 (II) ion and barbituric or thiobarbituric acid have been studied[23] in water, methanol, ethanol and acetone. They absorb maximally at 400 nm and adhere to Beer's law. The colorimetric determination of NO_2^- is accomplished[24] *via* conversion of 2-thiobarbituric acid to 2-thiovioluric acid (λ_{max} 570 nm).

Intense coloured complexes are reported[25] when heavy metal ions interact with 2-thiobarbituric acid and its derivatives. The complexes of copper[26], bismuth[27] and iron[28] with 2-thiobarbituric acid have been characterised (Table II) and the metals are determined colorimetrically. Rh (III) and Pd (II) have been determined[29] gravimetrically with the acid. The reactions are sensitive and specific. The colour reactions of trithiobarbituric acid with Cu (I), Cu (II), Ag (I), Hg (I), Hg (II) and Pd (II) have been used[30] in detection of traces of these metal ions.

TABLE I

Colour reactions of some barbituric acids

Acid	Reaction with	
	Copper (II)	Cobalt (II)
Barbituric	Purple solution	Blue solution
Thiobarbituric	Green solution	Purple precipitate
Nitrobarbituric	Blue solution	Green solution

TABLE II

Characteristics of metal complexes with 2-thiobarbituric acid

Characteristic	Cu (II)-complex	Bi (III)-complex	Fe (III)-complex
Colour	Yellow	Yellow	Rose red
λ_{max} (nm)	400	390	380
ϵ_{max}	6350	26000	..
Medium for complexation	0·1 N H_2SO_4	1 N HNO_3	pH 2·7–3·0
Composition (metal : ligand)	1 : 4	1 : 3	1 : 3
Range of determination	0·5–12 ppm	..	4–40 ppm
Interferences	Ag, Hg (II), Sn (II), Bi (III), Fe (III) Pd (III)	Cu (II), Ag, Cd (II), Hg (II), Sn (II), Sb (III)	Cu (II), Ag, Hg (I), Hg (II), Sn (II), Bi (III), Cr (III), Co (II)

2-Thiobarbituric acid has been widely used for photometric determination of 2-deoxy sugars, aldehydes, ketones, uronic acids and some other organic compounds. Periodate oxidation of deoxy sugars gives malonaldehyde which reacts with 2-thiobarbituric acid to give pink dye suitable for colorimetric determination[31,32]. In a modified procedure[33], the bisulphate addition compound of the aldehyde was isolated first before its reaction with the acid. A similar technique was adopted earlier in the detection of butter fat oxidation[34]. 2-Thiobarbituric acid in concentrated HCl is used in colorimetric estimation of ketoses[35], *viz.*, fructose and sucrose. The absorbance is measured at 432 nm and the optimum range of determination is 0·02–0·2 micromoles. It is also used in colorimetric determination of vitamin A aldehyde[36] in various tissues and sialic acid[37,38], an important constituent of human semen. The sialic acid is oxidised by $NaIO_4$ in concentrated H_3PO_4 and the oxidation product is coupled with 2-thiobarbituric acid to produce a coloured compound extractable in cyclohexanone and suitable for photometric determination. The method is specific and applied[39] in direct analysis of the acid in tissue hydrolyzates.

Uronic acids except galactouronic acid react with thiobarbituric acid after treatment with cold alkali giving a rose red dye (λ_{max} 548 nm) suitable for their colorimetric determination[40]. Galactouronic acid[41] reacts with the compound in 1 : 1 H_2SO_4 — CH_3COOH medium to produce yellow coloured species (λ_{max} 400 nm).

Sorbic acid[42] asd quinic acid[43] are oxidised with $K_2Cr_2O_7 - H_2SO_4$ and $HIO_4 - H_2SO_4$ respectively and the resulting malondialdehyde is coupled with 2-thiobarbituric acid to give red dye. For determination of sorbic acid, the absorbance of the dye is measured at its λ_{max} 535 nm while for quinic acid, the dye (λ_{max} 550 nm) is extracted into cyclohexanone before its absorbance is measured. The determination of 5–200 μg/ml of formic acid[44] depends on its reaction with thiobarbituric acid in acidic medium to produce a chromophore (λ_{max} 450 nm).

Vanillin is condensed with barbituric acid and the product is used[45] as an acid-base indicator (yellow in acid and red in alkali). Azo-compound obtained by interaction of 2-amino-5-chlorophenyl-6-sulphonic acid with barbituric acid is used[46] for luminescent determination of Mg (II). Ten-fold excess of Cu (II) and Ga (III) and 100-fold excess of Hg (II), In (III), Gd (III), Ge (IV), Sn (II), Sn (IV), Sb (III) and Co (II) do not interfere. Fe (III) reacts with [5-hydroxy-2-(1, 3)-indandione-2-yl] barbituric acid dioxime[47] in 1 : 2 ratio (λ_{max} 370 nm; ϵ 890) at pH 4·2. Even 2–3 fold excess of Cu (II), Cr (III), Co (II) and Ni (II) interfere.

Malonaldehyde is condensed with barbituric acid to produce dihydrobarbituric acid trimethyne dye suitable for photometric determination of magnesium[48] (λ_{max} of the complex 540 nm). The method is applied to determine the metal in aluminium alloys by masking the interfering ions by KCN or NaF.

Uranil-N, N-diacetic acid-5-bis (carboxymethylaminobarbituric acid) is used for amperometric determination of Be[49] ($\geqslant 1 \cdot 1 \times 10^{-4}$ M), In[50] ($\geqslant 6 \cdot 4 \times 10^{-6}$ M) and Ga[50] ($1 \cdot 4 \times 10^{-5}$ M) using a graphite electrode. The complexes contain the reactants in 1 : 1 ratio. In determination of Be, the interference due to Cu, Mg, Ca, Zn, Al, Mn (II), Fe (II), Fe (III), Co (II) and Ni (II) is eliminated by EDTA. Ga (III) can be determined in metallic aluminum and with indium, simultaneously. Stability constants (potentiometric) of indium and gallium complexes are 12·88 and 12·05 (at 25° C) respectively.

The reactions between uranil diacetic acid and several metal ions like Cu (II), Cd (II), Ce (IV), UO_2 (II), Fe (II), Fe (III) and Ni (II), in acidic or neutral medium, have been investigated[51-53]. The equimolar complexes of the ligand with Cu (λ_{max} 775 nm), Cd (λ_{max} 265 nm), UO_2 (λ_{max} 375 nm) and Ni (λ_{max} 390 and 635 nm) follow Beer's law and are used for absorptiometric determination of the metals in minerals and coinage metals.

It is evident from the foregoing work that the reactions of barbituric acids are very potential in several useful detections and determinations. The acids also serve as sensitive and many a time quite selective reagents for determination of metals.

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1. Bolton, W., *Acta Cryst.*, 1963, 16, 166.
2. Naves, R. G. and Strickland, B., *Chemistry*, 1974, 47 (3), 15.
3. Mackney, A. W. and Reynolds, T. M., *J. Council Sci. Ind. Res.*, 1930, 11, 333.
4. Tishchenko, V. E. and Koshkin, N. V., *J. Appl. Chem. (U.S.S.R.)*, 1934, 7, 1307.
5. Koshkin, N. V. and Tokarev, U. P., *Ibid.*, 1936, 9, 171.
6. Asmus, E. and Garschagen, H., *Z. Anal. Chem.*, 1953, 139, 81.
7. —, and Kurandt, H. F., *Ibid.*, 1956, 149, 3.
8. Nielsch, W. and Giefer, L., *Ibid.*, 1960, 171, 401.
9. Asmus, E. and Papenfuss, O., *Ibid.*, 1962, 185, 201.
10. — and Garschagen, H., *Ibid.*, 1953, 138, 404.
11. — and —, *Ibid.*, 1953, 138, 414.
12. Seiffarth, K. and Ardelt, H. W., *Z. Chem.*, 1963, 3, 430.
13. Baresel, D. and Klaus, J., *F. Z. Anal. Chem.*, 1970, 249, 234.

14. Pelletier, O. and Campbell, J. A., *J. Pharm. Sci.*, 1962, **57**, 594.
15. Tanabe, V. and Hata, K., *Ann. Rept. Fac. Pharm. Kanazawa Univ.*, 1956, **6**, 7.
16. Kozelka, F. L. and Tatum, H. J., *J. Pharmacol.*, 1937, **59**, 54.
17. Liberalli, C. H., *Rev. Quim. Farm.*, 1938, **3**, 153.
18. Lobanov, V. I., *Zh. analit. Khim.*, 1965, **20**, 881.
19. Danelsson, B., *Svensk Farm. Tidskr.*, 1954, **58**, 511.
20. —, *Ibid.*, 1951, **55**, 125.
21. Budde, H., *Apoth Ztg.*, 1934, **49**, 295.
22. Kloeking, H. P., *Z. Chem.*, 1962, **2**, 310.
23. Florica, P. and Constanta, P., *Rev. Roum. Chim.*, 1968, **13**, 473.
24. Krylov, V. P. and Drozdov, N. S., *Khim. Nauka i Prom.*, 1958, **3**, 838.
25. Shimotani, M., *Ann. Rept. Fac. Pharm. Kanazawa Univ.*, 1952, **2**, 76.
26. Halina, S. T., *Mikrochim. Acta*, 1969, **4**, 715.
27. —, *Ibid.*. 1969, **4**, 718.
28. —, *F. Z. anal. Chem.*, 1968, **234**, 414.
29. Currah, J. E., McBryde. W. A. E., Cruikshank, A. J. and Beamish, F. E., *Ind. Eng. Chem. Anal. Ed.*, 1946, **18**, 126.
30. Pavodini, T. and Gamlarin, F., *Anal. Chim. Acta*, 1949, **3**, 180.
31. Waravedkai. V. S. and Saslaw, L. D., *J. Biol. Chem.*, 1959, **234**, 1945.
32. —, *Biochim. et Biophys. Acta*, 1957, **24**, 439.
33. —, *J. Org. Chem.*, 1957, **22**, 843.
34. Biggs. D. A. and Bryant, L. R., *Can. J. Technol.*, 1953, **31**, 136.
35. Percheron, F., *Bull. Soc. Chim. France*, 1963, p. 1684.
36. Futterman, S. and Saslaw, L. D., *J. Biol. Chem.*, 1961, **236**, 1652.
37. Warren, L., *J. Clin. Invest.*, 1959, **38**, 785.
38. —, *J. Biol. Chem.*, 1959, **234**, 1971.
39. Neukom, N., *Chimia*, 1960, **14**, 165.
40. Zimmermann, R., Bock, W. and Taeufel, K., *Z. anal. Chem.*, 1962, **186**, 350.
41. Steigmann, A., *Sci. et indus. phot.*, 1958, **29**, 260.
42. Davidek, J. and Frimlova, E., *Vyskoke. skoly. Chem. Technol. Praze Potravinarska Technol.*, 1962, **6**, 47.
43. Mesnarel, P. and Devaux, G., *Compt. Rend.*, 1963, **256**, 1551.
44. Barker, S. A. and Somers, P. J., *Carbohydrate Research*, 1966, **3**, 220.
45. Pavolini, T., *Revista. ital. Essenze-e-profumi*, 1930, **12**, 93.
46. Serebryakova, G. V., Lukin, A. M. and Bozhevol'nov, E., *Zh. analit. Khim.*, 1963, **18**, 706.
47. Kucher, E., *Acta Fac. Rerum Natur. Univ., Comenianae Chim.*, 1970, **14**, 23.
48. Asmus, E. and Ortlepp, W., *F. Z. anal. Chem.*, 1973, **266**, 343.
49. Gallai, Z. A. and Zveryaeva, I. V., *Zh. anal. Khim.*, 1971. **26**, 2340.
50. Busev, A. I., Gallai, Z. A. and Zveryaeva, *Zavod Lab.*, 1974, **40**, 514.
51. Poch, M. M., *Acta cient compostelana*, 1972, **9**, 81.
52. Martinez, F. B. and Poch, M. M., *Ibid.*, 1973, **10**, 45.
53. — and —, *Microchim. J.*, 1974, **19**, 373.

DECOMPOSITION OF NITROUS OXIDE ON LANTHANUM-NICKEL OXIDE

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INTRODUCTION

THE decomposition of nitrous oxide over simple metal oxides has been studied in detail[1-6], from the kinetic as well as methanistic viewpoint of the reaction. In all the investigations mentioned above, only the initial stages of the reaction were considered and it appears that no satisfactory attempt has been made to study the kinetics of this reaction completely. In the present study. the decomposition of nitrous oxide has been carried out on a ternary oxide system like La_2NiO_4, to evaluate the suitability of this compound as a catalyst in oxidation reactions, in view of the favourable structural properties reported for this compound[7-9].

The decomposition of nitrous oxide has been studied over lanthanum nickel oxide in a static reactor (volume 125 cc) with an electromagnetically operated gas recirculation assembly. The course of the decomposition of N_2O was followed by following the changes in total pressure as a function of time. The N_2O decomposition was studied in the temperature range of 300°–400° C.

The catalyst was degassed for 6 hours at 420° C and at pressures $< 10^{-5}$ Torr and then treated with oxygen (10 cm pressure) for 10 hours at the reaction temperature. The pretreated catalyst was then subjected to evacuation for 2 minutes to remove excess oxygen and then decomposition of N_2O was studied by admitting 50–100 Torr of N_2O.

The catalyst La_2NiO_4 was prepared by firing a mixture of La_2O_3 (Indian Rare Earths Ltd., 99·9% pure) and nickel dimethylglyoxime complex. X-ray powder diffraction patterns of this sample agreed well with those cited in literature for this compound[10]. The nitrous oxide (Medical grade), oxygen and nitrogen (supplied by Indian Oxygen Limited) and helium were purified by standard procedures[11].

The catalyst had a BET (N_2) surface area of 2·7 m^2 gm^{-1}. The kinetic data for the decomposition of nitrous oxide was analysed using the integral form of the expression.

$$\frac{-dP_{(N_2O)}}{dt} = kp_{(N_2O)} \qquad (1)$$

and this gave satisfactory analysis of the data. The first order plots were linear with the psuedo rate constant dependent upon the initial pressure of nitrous oxide used. The results are summarized in Table I.

TABLE I

Kinetic parameters for the decomposition of N_2O on La_2NiO_4

Temp. (° C)	$(P_{N_2O}) \rightarrow 0$ Torr	K_{app} Min^{-1}	n
300	50 100	$4·386 \times 10^{-3}$ $2·303 \times 10^{-3}$	$-0·92$
320	50 100	$5·527 \times 10^{-3}$ $4·386 \times 10^{-3}$	$-0·35$
340	50 100	$13·16 \times 10^{-3}$ $8·064 \times 10^{-3}$	$-0·07$
360	50 100	$15·79 \times 10^{-3}$* $17·40 \times 10^{-3}$	0
370	100	$21·05 \times 10^{-3}$	
	50	..	
400	100	$30·40 \times 10^{-3}$	

* at 355° C.

The influence of the initial pressure of nitrous oxide upon the kinetics of decomposition was derived from the rate law.

$$r = kp^n \qquad (2)$$

where 'n' is the order of reaction.

The apparent order of the reaction for the decomposition of N_2O varies from $-1·0$ to 0 as the temperature of the reaction is increased from 300° to 400° C.

The energy of activation for the reaction was evaluated from Arrhenius plots. The energy of activation for the initial stages of the reaction in the temperature range of 300°–350° C at the initial pressures of N_2O of 50 Torr and 100 Torr are respectively 1·5 and 2·3 kcal/mole. The energy of activation for the reaction further reduces to 1·0 kcal/mole in the temperature range of 360°–400° C where the order of the reaction becomes zero.

Considering the presence of F-centers on a complex oxide of the type A_2BO_4, the decomposition is initiated by the adsorption of N_2O on a F-center, which further undergoes the decomposition. The results could be explained assuming the rate determining step to be either the surface decomposition of nitrous oxide or the adsorption of nitrous oxide.

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1. Amphlett, C. B., *Trans. Faraday Soc.*, 1954, **50, 273.**
2. Winter, E. R. S., *J. Catal.*, 1970, **19**, 32.
3. Gay, and Tompkins, F., *Proc. Roy. Soc.*, Sec. A., 1966, **293**, 19.
4. Cremer, E. and Marschall, E., *Montager*, 1951, **82**, 840.
5. Winter, E. R. S., *J. Catal.*, 1969, **15**, 144.
6. Read, J. F., *Ibid.*, 1973, **28**, 428.
7. Foex, M., *Bull. Chem. France*, 1961, p. 109.
8. Smolenski, G. A., Yudin, V. M. and Sher, E. S., *Soviet Phys. Solid State* (*ENG*), 1963, **4**, 2452.
9. Rao, C. N. R. and Ganguly, P., *Mat. Res. Bull.*, 1973, **8**, 405.
10. Rabenau, A. and Eckerlin, P., *Acta Cryst.*, 1958, **11**, 304.
11. Dodd, R. E. and Robinson, P. L., *Experimental Inorganic Chemistry*, Elsevier Publishing Co., 1957, Chapter II.

VIOLURIC ACIDS AS ANALYTICAL REAGENTS

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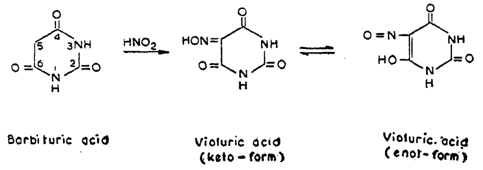
ABSTRACT
Analytical applications of violuric acids have been reviewed.

THE studies on violuric acids, which can be prepared by action of nitrous acid on barbituric acids, were started in 1863 when Baeyer[1,2] carried out experiments to elucidate the structural relationship between uric acid and its decomposition products, one of which is violuric acid. Infra-red studies[3,4] show that they exist in keto-enol forms. Techow[5], Andreasch[6,7], Hartley[8], Hantzsch *et al.*[9,10] and Meek and Watson[11,12] studied the colour, solubilities and absorption spectra of several violuric acids and their organic and inorganic salts from the point of view of colour in relation to chemical constitution. Colour intensity of the organic salts of the acids was related[13] to the strength of the corresponding bases. Later on thiovioluric acids and their metallic salts were prepared and characterised[14-17]. Numerous papers have appeared suggesting the use of the compounds as analytical reagents. In the present article, their analytical applications are reviewed.



Violuric acid has been used as a spraying reagent[18-20] in column and paper chromatographic analysis for identifying the separated elements. Alkali and alkaline earth metals form characteristic coloured salts with the acid having different solubility in anhydrous dimethylformamide. Based on this observation, sodium present in blood serum is determined spectrophotometrically[21]. Violuric acid has also been used as an indicator in complexometric determination of copper[22] in presence of several cations at pH 5–8. Palladium is detected and gravimetrically determined[23-26] as violurate complex, $Pd(C_4H_2N_3O_4)_2$ in the presence of many fold excess of Cu (II), Mn (II), Fe (II), Fe (III), Co (II) and Ni (II). The violurate starts decomposing at 340° C and metallic palladium results at 700° C. The sensitivity for the detection of the metal and limit of dilution are 0·2 μg/ml and 1 : 150,000 respectively. Violuric acid gives violet colour with carbonates and other alkaline salts of group I metals. As low as 0·3 μg Cu (II) and 0·1 μg Fe (II) can be detected with 10–15 fold excess of the acid when the metals give yellow and blue colours respectively at dilution of about 1 : 200,000. Cu (II)[27], Fe (II)[28], Co (II)[29-31] and Ni (II)[30] are spectrophotometrically determined with the compound; many diverse ions do not interfere. The characteristics of the complexes are summarised in Table I. The shapes and colours of the crystals of insoluble violuric acid salts of Li, Rb, Cs, Ca, Sr, Ba, Zn, Cd, Tl, UO_2 and Co have been investigated and a scheme proposed for their qualitative analysis. Zinc violurate is used[33] as a specific reagent for detection of Cu (II) in neutral solution (limit of identification : 3 μg).

TABLE I

Characteristics of metal complexes with violuric acid

Characteristic	Violuric acid complex with			
	Cu (II)	Fe (II)	Co (II)	Ni (II)
Colour ..	Yellow	Blue	Yellow	Yellow
λ_{max} (nm) ..	415	619	368	360
pH range for complexation ..	4·5–5·5	3·9–9·0	7·5	7·5
Adherence to Beer's law (ppm) ..	..	0·06–2·47	6–20	..
Composition (metal : ligand)	1:2	1:2	1:3	1:2

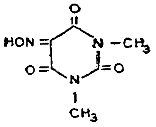
Alkali and alkaline earth metal salts are spectrophotometrically titrated with 1, 3-dimethylvioluric acid[34]. The determination is carried out in nitrogen atmosphere at 538 nm. Diphenylvioluric acid[35-37] forms complexes with metal ions, *viz.*, Cu (II), Ag (I), Au (III), Ca (II), Ba (II), Hg (II), Al (III), Ce (IV), UO_2 (II), Th (IV), Sn (II), Ti (IV), Zr (IV), Bi (III), Cr (III), Fe (II), Fe (III), Co (II), Ni (II) and Pt (IV). The com-

TABLE II

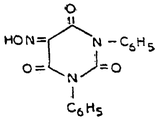
Characteristics of metal complexes with thiovioluric acid

Metal ion	Characteristics of metal complexes				
	Colour	λ_{max} (nm); ϵ_{max}	pH for const. absorbance	Composition (M: ligand)	Beer's law range (ppm)
Cu (II) ..	Yellow	395; 8,000	4·5–7·5	1:1	0·76–4·07
Fe (II) ..	Greenish blue	640–660; 24,000	7·8–9·7	1:3	Upto 1·78
Co (II) ..	Yellow	425; 58,000	7·0–9·3	1:3	Upto 1·30
Ni (II) ..	Yellow	375–395; 23,000	8·0–9·1	1:2	Upto 3·0
Ru (III) ..	Pinkish	520–540; 22,000	4·5–6·0	1:2	Upto 5·36
Rh (III) ..	Yellow	395; 18,000	5·6–7·2	1:3	Upto 3·60
Os (VIII) ..	Reddish violet	440–480	5·0–6·5	1:2	Upto 15·22
Ir (IV) ..	Yellow	395 and 425; 11,800	8·0–9·0	1:3	Upto 12·35
Pt (IV) ..	Yellow 6,000	395; 6,000	4·4–5·5	1:2	4·6–23·0

pound finds use as a developer in inorganic paper chromatography.

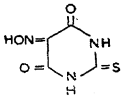


1, 3 Dimethylvioluric acid

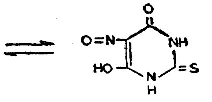


Diphenylvioluric acid

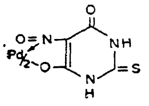
Similar to the use of violuric acids, the thio-derivatives are also employed for the purpose. Coloured metal complexes of thiovioluric acid[31,38-41] (Table II) are suitable for spectrophotometric determination of the metals. Palladium-thioviolurate[42] precipitated at pH 0·3–4·0 is thermally stable and dried at 110–130° C for gravimetric determination of the metal. With 10 mg of palladium precipitated at pH 0·3, there is no interference from the following ions in the amounts (mg) shown: Au (III) 22·8, Zn (II) 60, Cd (II) 50, Hg (II) 105, Tl (I) 30, Pb (II) 140, Ti (IV) 100, Zr (IV) 90, Th (IV) 20, As (V) 80, Sb (V) 25, Se (VI) 40, UO_2 (II) 70, Mn (II) 60, F^- 60, CH_3COO^- 100, $C_2O_4^{2-}$ 75, $C_4H_4O_6^{2-}$ 110, BO_3^{3-} 50, PO_4^{3-} 50 and citrate 100; Cu (II) 65, Bi (III) 25 and Ru (III) 20 can be masked with 100 mg of tartrate or citrate. Diphenylthiovioluric acid (or its ammonium salt) has been used[43-48] to quantitatively precipitate and estimate copper (II)[41,42], iron (II), iron (III)[45-46] and nickel (II)[47] in the pH range 7·2–8·0, 4·9–5·6, 4·9–5·8 and 4·0–5·8 respectively. The metal complex obtained in each case is dried in the sintered crucible at 110–115° C and weighed.



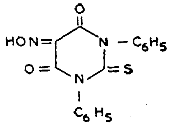
Thiovioluric acid (Oxamino-ketonic form)



Thiovioluric acid (Nitroso-enolic form)



Palladium-thioviolurate complex



Diphenyl Thiovioluric acid

The results are accurate within 0·5% error. Iron, cobalt, ruthenium and palladium complexes with the ligand have also been investigated spectrophotometrically[37, 49-50]. All the complexes are extractable in chloroform and used in extractive photometric determination of the metals. Their characteristics are summarised in Table III. In a titrimetric procedure developed[51] for estimating copper, the metal complex is precipitated from copper salt solution by the ligand, filtered and the residue treated with NaOH to give the sodium salt. The mixture is acidified with HCl and the liberated diphenylthiovioluric acid estimated by bromate titration and thus copper indirectly determined. In nitrogen atmosphere, lead is amperometrically titrated[52] with this ligand. The composition of the precipitate at the equivalence point corresponds to 1 : 2 (Pb : ligand). In a recent work[53], thin layer chromatographic technique is used in separation of diphenylthiovioluric acid-metal chelates.

TABLE III

Characteristics of metal complexes with diphenylthiovioluric acid

Characteristic	Diphenylthiovioluric acid complex with			
	Fe (II)	Co (II)	Ru (III)	Pd (II)
Colour	Greenish blue	Yellow	Red	Yellow
λ_{max} (nm)	630–640	415	520	425
ϵ_{max}	18,100	51,400	22,300	30,000
pH for constant absorbance	4·5–6·5	5·8–7·5	4·9–6·7	1·0–3·5
Beer's law range (ppm)	Upto 3·27	Upto 2·92	Upto 5·60	Upto 4·96
Composition (metal : ligand)	1 : 3	1 : 3	1 : 2	1 : 2

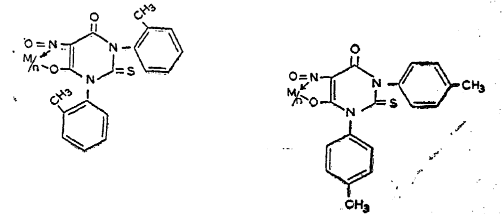
Diortho-tolylthiovioluric acid forms blue complex with Fe^{2+} (λ_{max} 660 nm) at pH 3–6. The reagent is used[54] in detection (1 part in 6 million parts of water) and colorimetric determination of the metal. Large number of inner metal complexes with di-*ortho*-[55] and di-*para*-tolylthiovioluric acids[56] have been prepared and characterised.

Stabilities of Fe (II) complexes formed with diphenyl- and (*o, m* and *p*)-ditolylthiovioluric acids have been compared[57]. Log K values of the chelates are related to pK_a values of the ligands (Table IV).

TABLE IV

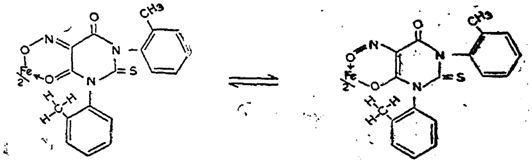
pK_a values of substituted thiovioluric acids and log K values of their Fe (II) chelates

Ligand/Iron (II)-complex with ligand	pK_a of the ligand	Log K of iron (II) complex
Diphenylthiovioluric acid ..	5·00	5·20
Di-*o*-toylthiovioluric acid ..	5·35	5·60
Di-*p*-tolylthiovioluric acid ..	5·60	5·80
Di-*m*-tolylthiovioluric acid	5·90	6·08



Metal complexes with di-*o*- and di-*p*-tolylthiovioluric acids, M = mono-, di, ter- or tetravalent metal, *n* = oxidation state of the metal.

Introduction of the methyl group in the side benzene ring enhances the basic character of the ligands and also the stability of the metal chelates. The effect of the methyl group is most pronounced when it is in the *meta* position ; in the *ortho* derivative (resonating structures shown below) the effect is minimum because of steric factors. Several substituted thiovioluric acids have also been used in detection of iron (II)[58] (sensitivity 0·05 μg on spot plate ; dilution limit 1 : 1,000,000), and gravimetric determination[59] of Ag (I), Fe (II), UO_2 (II) and *organic bases aniline, toludines* (*o-*, *m-* and *p-*) and naphthaylamines (α- and β-). Stability constants of bivalent metal complexes with thiovioluric and diphenylthiovioluric acids have been determined potentiometrically[60] in 50 and 75% dioxan media, respectively (Table V). The order of stability constants for the complexes is : Mn < Ni < Cu > Zn.



Resonating structures of iron complex with di-*o*-tolylthiovioluric acid.

TABLE V

Stability constants of bivalent metal complexes with thiovioluric acid and diphenylthiovioluric acid

Ion	Thiovioluric acid (50% dioxan)		Diphenylthiovioluric acid (75% dioxan)	
	Log K_1	Log K_2	Log K_1	Log K_2
H ..	4·93	..	5·38	..
Cu (II) ..	4·44	3·91	5·13	4·30
Ni (II) ..	3·82	4·30	3·80	5·10
Pb (II) ..	3·44	..	3·84	..
Zn (II) ..	3·21	..	3·37	..
Mn (II)	2·93	..	3·09	..
Mg (II) ..	2·63	..	2·79	..

The stability of lead complexes is intermediate between Mn (II) and Ni (II) while magnesium forms the least stable complexes.

1. Baeyer, A., *Ann.*, 1863, **127**, 199.
2. —, *Ibid.*, 1864, **130**, 140.
3. Taylor, M. E., *Ph.D. Thesis*, Univ. of Washington, 1960.
4. Jons, R. N., "Infra-red spectra of organic compounds," *Can. Natl. Res. Compt. Bull.*, 1959, No. 6.
5. Techow, W., *Ber. Deut. Chem. Ges.*, 1894, **27**, 3082.
6. Andreasch, R., *Monatsh. Chem.*, 1895, **16**, 17.
7. —, *Ibid.*, 1900, **21**, 286.
8. Hartley, W. N., *J. Chem. Soc.*, 1905, **87**, 1791.
9. Hantzsch, A. and Ishwerwood, P. C. C., *Ber. Deut. Chem. Ges.*, 1909, **42**, 986.
10. — and Issaias, B., *Ibid.*, 1909, **42**, 1000.
11. Meek, D. B. and Watson, E. R., *J. Chem. Soc.*, 1915, **107**, 1567.
12. — and —, *Ibid.*, 1916, **109**, 553.
13. Ghatak, N. and Dutt, S., *J. Indian Chem. Soc.*, 1928, **5**, 665.
14. Lal, R. and Dutt, S., *Proc. Nat. Inst. Sci. Ind.*, 1937, **3**, 337.
15. Prakash, S. and Dutt, S., *Ibid.*, 1938, **8**, 29.
16. Dass, I. N. D. and Dutt, S., *Proc. Indian Acad. Sci.*, 1938, **8**, 145.
17. Gambhir, I. R. and Singh, R. P., *Ibid.*, 1946, **23 A**, 330.
18. Hahn, H. V., Erlenmeyer, A. and Sorki, E., *Helv. Chim. Acta*, 1956, **39**, 341.
19. Weiss, A. and Fauab, S., *Ibid.*, 1954, **37**, 1253.
20. Hahn, H. V., Prijs, B. and Erlenmeyer, A., *Ibid.*, 1956, **39**, 341.
21. Muraca, R. F. and Bonsack, J. P., *Chemist Analyst*, 1955, **44**, 38.
22. Solodovnikov, P. P., *Zh. Anal. Khim.*, 1963, **18**, 1026.
23. Ziegler, M. and Glemsen, O., *Mikrochim. Acta*, 1956, p. 1152.
24. — and —, *Ibid.*, 1956, p. 1515.
25. Bogonostsev, A. S., *Izv. Vysshikh Uchebn Zavedenii Khim i Khim Tekhnol*, 1963, **6**, 1046 ; *C.A.*, 1964, **61**, 1263 *d*.
26. —, *Ibid.*, 1964, **7**, 15 ; *C.A.*, 1964, **61**, 4944 *g*.
27. Ziegler, M., *Z. Anal. Chem.*, 1958, **164**, 387.
28. Cerny, P., *Chem. Listy*, 1957, **51**, 735.
29. Shiro, H. and Koji, M., *Bunsek. Kagaku*, 1969, **18**, 1017.
30. Ershova, L. V., Irqnov, V. M. and Busev, A. I., *Zh. Anal. Khim.*, 1974, **29**, 1367.
31. — and Noskov, V. V., *Ibid.*, 1971, **26**, 2406.
32. Vanlingten, J. W. L. and Velthuyzen, H. V., *Mikrochim. Ichnoanal Acta*, 1964, **5**, 759.
33. Ueiwang, K., *J. Chines. Chem. Soc.*, 1950, **17**, 164.
34. Taylor, M. E. and Robinson, R. J., *Anal. Chem.*, 1962, **34**, 533.
35. Singh, R. P., *Curr. Sci.*, 1955, **24**, 208.
36. —, *J. Sci. Ind. Res.*, 1956, **15**, 245.
37. — and Purshottam, V., *Proc. Symposium on the Chem. of Coord. Compounds*, Part III, Agra (India), 1959, p. 262.
38. Chawla, R. S., *Ph.D. Thesis*, Delhi University, Delhi, 1970.
39. —, and Singh, R. P., *Mikrochim. Acta*, 1970, p. 332.
40. Ershova, L. V., *Zavod. Lab.*, 1972, **38**, 905.
41. Chawla, R. S. and Singh, R. P., *J. Indian Chem. Soc.*, 1975, **52**, 169.
42. —, Dutt, Y. and Singh, R. P., *Talanta*, 1969, **16**, 119.
43. Singh, R. P., *Curr. Sci.*, 1955, **24**, 208.
44. — and Shankar, V., *J. Indian Chem. Soc.*, 1955, **32**, 557.
45. —, *Curr. Sci.*, 1956, **25**, 59.
46. —, *J. Indian Chem. Soc.*, 1959, **36**, 57.
47. —, *J. Sci. Ind. Res.*, 1960, **19 B**, 468.
48. —, *Ph.D. Thesis*, Delhi University, Delhi, 1960.
49. Malik, L. C. and Singh, R. P., *J. Indian Chem. Soc.*, 1956, **33**, 335.
50. Chawla, R. S., Singh, R. P. and Trikha, K. C., *Talanta*, 1971, **18**, 1245.
51. Singh, R. P. and Banerjee, N. R., *Curr. Sci.*, 1960, **29**, 223.
52. Banerjee, N. R. and Singh, R. P., *J. Indian Chem. Soc.*, 1963, **40**, 175.
53. Johri, K. N. and Mehra, H., *Chromatographia*, 1975, **8**, 244.
54. Shivahare, G. C. and Singh, R. P., *Univ. Raj. Physical Sci. Studies*, 1956, p. 23.
55. Singh, R. P., *J. Indian Chem. Soc.*, 1959, **36**, 198.
56. —, *Ibid.*, 1960, **37**, 569.
57. — and Banerjee, N. R., *Ibid.*, 1960, **37**, 713.
58. —, *Ibid.*, 1960, **37**, 447.
59. Bhargava, P. N., Veerabhadriah, N. and Satyanarayana, B., *Ibid.*, 1957, **34**, 889.
60. Chawla, R. S. and Singh, R. P., *Microchem. J.*, 1973, **18**, 646.

LETTERS TO THE EDITOR

STUDIES OF STANDARD POTENTIAL OF COBALT-COBALT PYROPHOSPHATE ELECTRODE

Introduction

SUITABLE electrodes reversible with respect to pyrophosphate ions have not been developed so far. A systematic and comprehensive study in this direction comprising the determination of E° (standard electrode potential) of the various electrode systems was considered desirable. The e.m.f. values of cobalt-cobalt pyrophosphate cells with and without transport and the potential of cobalt-cobalt pyrophosphate electrode are worth reporting. The experiments were similar to those of Pearce and Ough[1] with minor modifications necessary to account for the colloidal nature of cobalt pyrophosphate as well as to show the presence of cobalt ions in the electrical double layer of cobalt pyrophosphate sol.

MATERIALS AND METHODS

Reagents :

All the reagents employed were of A.R. quality. Mercury employed for preparing the amalgam as well as mercurous chloride was triply distilled.

Preparation of the Complex :

Solutions of cobalt nitrate and pyrophosphoric acid [conc. 1·0 M each] were mixed in the molar ratio of 1 : 2 and allowed to stand for about 2 hours. The precipitate obtained was filtered, washed several times with water and then dried at 90° C. The sodium amalgam electrode was prepared as described in literature.

Cobalt Electrode :

A thin film of cobalt was deposited on a platinum foil (area 1 sq. cm) by the electrolysis of a solution containing cobalt sulfate (56%), boric acid (3%) and NaCl (2%). A current of 50 mA was passed for 4 hours from a 4 V accumulator.

RESULTS AND DISCUSSION

1. Measurement of e.m.f. of cells with and without transport :

Cells of the type I (with transport) and II (without transport) were set up. The electrode vessels employed were of the type used by Harned[2]. A small bed of glass wool was placed over the narrow mouth of the constriction provided in the short limb of the vessel and the cobalt pyrophosphate covered the cobalt surface of the electrode completely. The cobalt electrode was then inserted into the powdered cobalt pyrophosphate. Air from the vessel was then removed by evacuation and replaced by hydrogen. Sodium pyrophosphate was introduced into the vessel under hydrogen pressure. For every concentration of sodium pyrophosphate two half cells were first set up, both were connected to salt bridges and short circuited for the attainment of equilibrium. The e.m.f. values are given in Table I. The following cells were set up for the studies :

$\mathrm{Co/Co\ (H_2P_3O_7),\ Na_4P_2O_7\ (0\cdot1\ m)\ |\ (0\cdot05\ m)}$
$\mathrm{Na_4P_2O_7,\ Co\ (H_2P_2O_7)_2/Co}$ I
$\mathrm{Co/Co\ (H_2P_2O_7)_2,\ Na_4P_2O_7\ (0\cdot1\ m)\ |\ NaHg\ |}$
$\mathrm{(0\cdot05\ m)\ Na_4P_2O_7,\ Co\ (H_2P_2O_7)_2/Co}$ II
$\mathrm{Co/Co\ (H_2P_2O_7)_2,\ Na_4P_2O_7\ Cx\ NaCl\ (Sat.)\ |}$
$\mathrm{NaCl\ (Sat.),\ Hg_2Cl_2/Hg}$ III

TABLE I

E.M.F. values of cobalt-cobalt pyrophosphate cells withI and without transport at 30° C

E (without transference) V	E_t (with transference) V
0·0350	0·0245
0·0290	0·0225
0·0365	0·0268
0·0355	0·0252

TABLE II

Potential of cobalt-cobalt pyrophosphate electrodes at 30° C

E.M.F. of the cell in volts			
Conc. of $Na_4P_2O_7$			
0·1	0·05 m	0·025 m	0·0125 m
0·5320	0·5280	0·5300	0·5220
0·5340	0·5270	0·5200	0·5195
0·5335	0·5275	0·5280	0·5180
0·5325	0·5285	0·5270	0·5170
0·5315	0·5300	..	..
..	0·5320	0·5260	0·5160
0·5315	0·5310	..	..
0·5330	0·5315	0·5275	..
E (Saturated Calomel electrode) V			
0·3458	0·3458	0·3458	0·3458
E, Co/Co $(H_2P_2O_7)_2$, V			
−0·3512	−0·3472	−0·3438	−0·3472

The cell of type III was used to determine the electrode potential of the pyrophosphate electrode at diffeernt conc. of $Na_4P_2O_7$. Eight such half cells were set up for each conc. of $Na_4P_2O_7$.

Calculations of Standard Potential[3] (E°) :

The value of E° of cobalt-cobalt pyrophosphate electrode was determined by the method employed by Lewis and Randoll[4]. The standard electrode potential for the cobalt-cobalt pyrophosphate electrode was found to be $-0 \cdot 367$ V.

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1. Pearce, J. N. and Ough, L. D., *J. Am. Chem. Soc.*, 1938, 60, 80.
2. Harned, H. S., *Ibid.*, 1929, 51, 416.
3. Glasstone, S., *An Introduction to Electrochemistry*, D. Van Nostrand, New York, 1962, p. 271.
4. Lewis, G. N. and Randall, M., *Thermodynamics*, McGraw-Hill Book, Co., Inc., New York, 1923, p. 334.

ACETATES OF COBALT AND COPPER AS SPRAYING REAGENTS FOR THE DETECTION OF SOME OPIUM ALKALOIDS

OPIUM and its alkaloids have been detected on thin layer plates by various spraying reagents, which were reviewed by Santavy[1]. Deniges reagent[2], iodine fuming and subsequent exposure to pyrrole vapours[3], have been reported as detecting agents for opium alkaloids.

In the present paper, detection of opium alkaloids, by aqueous solutions of acetates of copper and cobalt, has been described. The developed thin layer plates are first sprayed with an aqueous solution of either copper acetate or cobalt acetate. Then both the plates have been subsequently sprayed with a saturated solution of ammonium thiocyanate in acetone. The second spray with the saturated solution of ammonium thiocyanate in acetone revealed the alkaloids as coloured spots. The colour of the spots is due to the formation of a complex of cobalt (II) thiocyanate and copper (II) thiocyanate with alkaloids.

Glass plates of size 20 × 20 cm were coated with silica gel G(E/M), approximately to a thickness of 300 μm and the plates after air drying, were activated at 110° C for one hour. Then the plates were cooled in a desiccator. Methanolic solution of morphine and chloroform solutions of codeine, narcotine, papaverine and thebaine were spotted on the plates by means of a micropipette. The plates were then developed in a solvent mixture of benzene : methanol (80 : 20). After the solvent front had moved to 10–12 cm, the plates were taken out of the solvent chamber and dried with a hair drier.

(*a*) The plates was sprayed with 5% aqueous solution of cobalt acetate and dried for 15 minutes and then sprayed with a saturated solution of ammonium thiocyanate in acetone. The alkaloids formed deep greenish blue spots on a very light blue background. Minimum detectable limits by this reagent system are 5 μg for morphine, narcotine, thebaine and papaverine, and 30 μg for codeine. It was observed that with the spray of 5% aqueous solution of cobalt acetate alone and subsequent hot air treatment, the alkaloids gave violet coloured spots on a white background.

(*b*) A second plate was sprayed with 5% aqueous solution of copper acetate and dried for 15 minutes as above. The plate was then sprayed with the saturated solution of ammonium thiocyanate in acetone, which revealed the alkaloids as chocolate brown coloured spots. Minimum limits of detection with this spray reagent are 5 μg for morphine, narcotine, papaverine and thebaine and 25 μg for codeine.

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1. Santavy, F., In : E. Stahl (Editor), *Thin Layer Chromatography*, A Laboratory Hand Book, Springer, New York, 1969.
2. Guven, K. C., and Aran, B., *Anal. Abstr.*, November 1973, 25, 3340.
3. Rama Rao, N. V. and Murthy, H. R. K., *J. ind. Acad. For Sci.* (13), 1974, p. 18.

NEW MERCURATED SULFONAMIDES AS POTENTIAL PESTICIDAL PROPERTIES

RECENTLY, sulfonamides which have wide-spread use as bactericide[1], have also been found to possess fungicidal[2] properties and a large number of them synthesised[3,4] and tested for their pesticidal behaviour. For such purpose, mercurated sulfonamides, substituted aniline bases treated with halo alkyl, arylalkyl or aryl sulfonyl chloride[5] and aryl sulfonamides[6] have been prepared. Guha-Sircar and Ismet Ali[7] also synthesised a series of mercurated sulfonamides and reported potent bactericidal properties for their compounds.

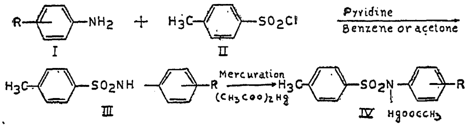
TABLE I

Nature of the synthesized compounds and their properties

Sl. No.	Nature of R	M.P. (°C)	YieldI/	Mercury percentage		Pesticidal properties				
				Found	Calculated	Antifungal			Antibacterial	
						A.N.	C.G.	E.S.	E.C.	S.A.
1.	H–	250	60	39·3	39·5	+	+	+	+	+
2.	2-methyl	225	58	38·0	38·5	+	+	+	+	+
3.	3-methyl	210	65	38·1	38·5	−	−	−	+	−
4.	2-methoxy	240	50	37·0	37·3	−	−	−	−	−
5.	3-methoxy	260	51	37·1	37·3	−	−	−	+	+
6.	4-ethoxy	205	64	37·0	37·2	++	++	+	++	++
7.	2-chloro	180	58	36·8	37·0	++	++	+	+	+
8.	3-chloro	260	55	36·7	37·0	+	+	+	+	+
9.	2-nitro	262	50	36·0	36·3	−	−	−	+	+
10.	3-nitro	275	55	36·7	36·3	++	++	++	+	+
11.	4-nitro	215	52	36·1	36·3	++	++	++	+	+
12.	2-methyl-4-chloro	247	50	35·6	36·0	++	+	+	++	−
13.	2-nitro-4-methyl	242	51	35·1	35·4	−	−	−	−	−
14.	2-methoxy-4-nitro	237	60	34·6	35·0	−	−	−	−	−
15.	3-nitro-6-methyl	236	58	35·1	35·4	++	++	+	++	+
16.	3-nitro-6-methoxy	230	61	34·8	35·0	++	+	+	+	−

(−) inferior to Zineb.
(+), (++) greater toxicity over Zineb to the extent of (0–50/) and (51–100/) respectively.

This approach was made use of by the author in her current studies towards development of new series of mercurated sulphonamide type of compounds using a wide range of aniline bases with $-CH_3$, $-OCH_3$, $-OC_2H_5$, $-Cl$, $-NO_2$ substituents, singly or in combination, and condensing them with *p*-toluenesulfonyl chloride. The following general reactions have been stipulated :



Mercurated sulfonamides were prepared by condensing 0·1 mole of each aniline base (I) with 0·1 mole of *p*-toluenesulfonyl chloride (II) in presence of 1 : 1 pyridine and benzene or acetone. The mixture was refluxed in water bath for four hours, after which the solvent was removed under reduced pressure. The N-(R-phenyl) *p*-toluenesulfonamide (III) so formed was recrystallised in ethanol and subjected to mercuration taking their equimolar ratio dissolved in ethanol, and dilute acetic-acid respectively. The reaction was completed under constant stirring and refluxing for one hour. The mercurated compound (IV) separating out was recrystallised from acetic-acid. The structure of the compound was established through acid hydrolysis and subsequent identification of the decomposition products. Their composition was verified by estimation of mercury[8] as mercuric sulfide.

Pesticidal properties of the compounds were tested by standard methods[9,10] against three fungi, namely, *Aspergillus niger* (A. N.), *Chetomium globossum* (C. G.) and *Rhizoctonia solani* (R. S.) and two bacteria *i.e., Escherichia coli* (E. C.) and *Staphylococcus aureus* (S. A.) at a concentration of 1×10^{-5} mole/ml of the compound dissolved in ethanol or acetic acid-ethanol mixture. The toxicity of the compounds was determined on these cultures by measuring the zone of inhibition and their relative toxicity was rated against 'Zineb' (Zinc-ethylenebis-dithiocarbamate), as a standard. The physical and pesticidal properties are tabulated in Table I.

It may be concluded that the compounds containing 4-ethoxy, 2-chloro, 3 and 4-nitro and 2-nitro-6-methyl have relatively strong dual purpose toxicity for plant pathogens and bacteria.

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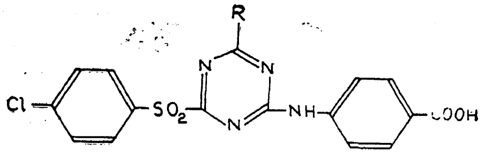
1. Budesiusky, Z., Bydzovsky, V., Prikryl, J. and Svab, J., *Ceskceslov. Farm.*, 1961, 10, 14 ; *Chem. Abstr.*, 1961, 55, 25972_e.
2. Angelo, J., Speziale, and Gino, J., *U.S. Pat.*, 1966, 3, 256. 148 ; *Chem. Abstr.*, 1966, 65, 6231_b.

3. Julian, B., Zygmunt, E., Zdzislaw, E., Bogumil, H., Wieslaw, S. and Jacek, S., *Prezemysl. Chem.*, 1961, 40, 561; *Chem. Abstr.*, 1962, 57, 12523_h.
4. Deo, K., *Japan Pat.*, 1959, p. 4328; *Chem. Abstr.*, 1963, 58, 10205_c.
5. Friendrich, M., *German Pat.*, 1959, p. 106238; *Chem. Abstr.*, 1961, 55, 14379_d.
6. Hans, J., Dietz, Wilhelm, B. and Frank, B., *Ger. Pat.*, 1956, p. 17905; *Chem. Abstr.*, 1961, 55, 1531_i.
7. Guha, S. S. and Ismet Ali, *J. Ind. Chem. Soc.*, 1951, 2, 28.
8. Lomhalt, S. and Christiansen, J. A., *Comp. Rend. Lab. Cerlberg.*, 1931, 10, 259.
9. Riber, A. J. and Riber, R. S., *Introduction to Research on Plant Diseases*, Academic Press, New York, U.S.A., 1936.
10. Mehrotra, G. and Dey, A. N., *J. Ind. Chem. Soc.*, 1973, 50, 793.

ANTIBACTERIAL ACTIVITY OF *s*-TRIAZINYL ARYL/ALKYL SULPHONES

p-AMINOBENZOIC acid derivatives have been found to be powerful local anaesthetics[1] and possess bacteriostatic property[2]. *s*-Triazine derivatives also possess therapeutic activity against malaria[3], cancer[4] and viral diseases[5]. We[7] have prepared 4′-(2-*p*-chlorophenylsulphonyl-4-aryl/alkylamino-*s*-triazine-6-yl)-aminobenzoic acids of the type (I) and tested for antibacterial activity.



(I)

where R = arylamino, alkylamino, etc.

The first chlorine of the cyanuric chloride was reacted with *p*-chlorothiophenol[6] at 0° C, leading to the formation of 2, 4-dichloro-*s*-triazine-6-yl *p*-chlorophenyl sulphide. The second chlorine was condensed with *p*-aminobenzoic acid at 30°–35° and the third chlorine at 80°–90° with different bases using dioxane as the solvent. The product was then oxidised to the corresponding sulphones[7].

Antibacterial Testing.—The following strains were used for testing the antibacterial activity:

A. Gram-positive bacterial strains like *Bacillus subtilis* and *Staphylococcus aureus.*

B. Gram-negative bacterial strains like *Escherichia coli,, Xanthomonas citri, Salmonella typhosa, Shigella shiga* and *Pseudomonas aeruginosa.*

Thirty-one sulphones were tested using dilution broth method *in vitro*. Loopful suspensions prepared from the above actively growing micro-organisms were inoculated into the nuterient broth containing different concentrations (800, 500, 400, 250, 200 and 100 μg/ml) of the sulphones and incubated for 24 hours at 37° C. The minimum inhibitory concentrations (MIC) were determined in μg/ml.

p-Bromoanilino, *p*-iodoanilino and 2, 4, 6-tribromoanilino derivatives were found to be most active as they inhibited the growth of Gram-positive bacteria at 200 μg/ml and Gram-negative bacteria at 400 μg/ml. The other substituents in the benzene ring of arylamino group like methyl, *m*-chloro, methoxy, ethoxy, nitro, hydroxy, sulpho, other heterocyclic derivatives like pyridylamino, morpholino, piperidino and alkyl-amino, arylalkyl-amino derivatives inhibited the growth of *B. subtilis* and *S. aureus* at 250 μg/ml; *E. coli, X. citri, P. aeruginosa* at 400 μg/ml; *S. typhosa* and *S. shiga* at 500 μg/ml.

o-, *m*- and *p*-carboxyanilino derivative inhibited the growth of Gram-positive bacteria at 250 μg/ml and Gram-negative bacteria at 500 μg/ml.

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1. Einhorn, A. and Uhfelder, E., *Ann.*, 1909, 371, 131.
2. Johnson, T. B. and Lane, F. W., *J. Amer. Chem. Soc.*, 1921, 43, 348.
3. Curd, F. H. S., Landquist, J. K. and Rose, F. L., *J. Chem. Soc.*, 1947, p. 154.
4. Burchenal, J. H., Johnston, S. F., Cremer, M. A., Webber, L. F., and Stock, C. C., *Proc. Soc. Exptl. Biol. Med.*, 1950, 74, 708.
5. CIBA Ltd., B. P. 754071, 1956. U.S.P. 2824088, 1958.
6. Drabek, J. and Skrobal, M., *Chem. Zvesti*, 1963, 17 (7), 482.
7. Goghari, M. H. and Parikh, A. R., *J. Indian Chem. Soc.*, 1976, 53, 207.

DRUGS ON PROTEIN BINDING OF TOLBUTAMIDE

SULPHONYL ureas have been reported to bind extensively to plasma proteins[1-2]. Displacement of sulphonyl ureas by number of acidic drugs has been well demonstrated in human serum[1,3] and in solutions of purified albumin[4-5]. The studies reported herein explore the influence of some drugs not reported earlier on protein binding of tolbutamide.

TABLE I

Binding of tolbutamide with Bovine serum albumin (moles/mole of protein) in presence of various molar concentrations of drugs

Drug	Concentration of drug				
	0	77×10^{-6} M	154×10^{-6} M	308×10^{-6} M	770×10^{-6} M
Paracetamol	2·34	1·98	1·40	0·98	0·51
Sulphathiazole	2·34	1·96	1·42	0·96	0·38
Sulphanilamide	2·36	2·01	1·73	1·36	0·90
Sulphadimidine	2·34	2·0	1·61	1·14	0·75
Analgin	2·34	2·35	2·32	2·34	2·32
Salicylamide	2·35	2·32	2·34	2·30	2·37
Phenobarbitone sodium	2·35	2·34	2·30	2·34	2·34
Chlorpheniramine maleate	2·35	2·33	2·38	2·31	2·40

Each value is an average of five readings. The dialysis system had the total volume of 12 ml and consisted of 24×10^{-6} M of albumin and 77×10^{-6} M of tolbutamide.

The binding studies were carried out by the technique of equilibrium dialysis. In a dialysis bag 1 ml of 2% Bovine serum albumin (Sigma Co.), 1 ml of tolbutamide (Hoechst, Bombay) solution of appropriate concentration and 1 ml of phosphate buffer (·067 M, pH 7·4) were placed, thus adjusting the volume to 3 ml. While testing the materials for their influence on protein binding of tolbutamide, 1 ml of the solution of the test drug of various concentrations was added to the bag instead of phosphate buffer. The solutions of albumin and drugs were all prepared in ·067 M phosphate buffer of pH 7·4. The mixture in the dialysis bag was dialysed against 9 ml of phosphate buffer (·067 M, pH 7·4) for 24 hours at 37° C. The bags were removed after 24 hours and the concentration of tolbutamide in the dialysate was estimated by the method of Spingler[7]. Control experiments using phosphate buffer instead of albumin solution were also performed. The total amount of drug bound to the protein was calculated from the difference between total amount of drug added and free drug concentration in the entire dialysis system. The binding of tolbutamide to protein is expressed in mole/mole of protein.

Of the compounds tested, paracetamol, sulphathiazole, sulphanilamide, sulphadimidine were found to interfere with protein binding of tolbutamide (Table I). The binding of tolbutamide with albumin was inhibited markedly with increasing concentration of these drugs. Sulphathiazole had the maximum inhibitory effect on protein binding of tolbutamide. It has been demonstrated that tolbutamide involves binding to 2 classes of sites on albumin[5,6]. The interference produced by the above drugs in binding of tolbutamide with protein suggests involvement of the common binding sites and relatively stronger affinities of the drugs than tolbutamide for the binding sites on protein. Especially, the sulpha drugs by virtue of possessing structural resemblance with tolbutamide can compete with tolbutamide for common binding sites. Sulphadiazine, however, failed to have any effect on protein binding of tolbutamide, presumably due to lower affinity than tolbutamide for binding with protein.

The interactions of the drugs with protein binding of tolbutamide bear clinical relevance since the displacement of the hypoglycemic agent from the carrier protein in presence of the competitor could aggravate hypoglycemic response and also reduce biological half life of the drug.

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1. Wishinsky, H., Glasser, E. J. and Perkal, S., *Diabetes,* 1962, 11 Suppl., 18.
2. Jackson, W. P. U., In *Oral Hypoglycemic Agents,* Editor, Campbell, G. D., Academic Press, London and New York, 1969.
3. Christenses, L. K., Hansen, J. M. and Kristensen, M., *Lancet.,* 1963, 2, 1298.
4. Buttner, H. and Portwich, F., *Klin. Wochschr.,* 1967, **45**, 225.
5. Judis, J., *J. Pharm. Sci.,* 1972, **61**, 89.
6. Crooks, M. J. and Brown. K. F., *J. Pharm. Pharmac.,* 1974, 26, 304.
7. Spingler, H., *Klin. Wochschr.,* 1957, **35**, 533.

EFFECTS OF THYROXINE ON THE ABUNDANCE OF CIRCULATING ERYTHROCYTES IN INTACT AND HYPOPHYSECTOMIZED MALE KILLIFISH, *FUNDULUS HETEROCLITUS*

EARLIER work has shown that erythrocytopenia is characteristic of hypophysectomized male *Fundulus heteroclitus,* and that it can be counteracted by administration of TSH to the fish[1]. But this effect of TSH was found to be due to the contamination of the hormone with LH which stimulated the testes; androgen is known for elevating the erythrocyte counts in the peripheral circulation of *Fundulus.* Further, hypothyroidism did not cause any significant change in the abundance of circulating erythrocytes in sea or fresh water adapted killifish. Occurrence of erythrocytopenia has also been reported in hypophysectomized *Poecilia latipinna,* and that it could not be alleviated by maintaining the fish for 4 weeks in L-thyroxine solution[2]. However, observations have been made that a significant decrease occurred in the number of erythrocytes in thyroidectomized rainbow trout[3], and that thyroxine treatment prevented anemia in captive sturgeons[4]. In mammals hypophysectomy-induced anemia has been attributed to decreased need for oxygen by the tissues when thyroid, or thyrotrophic and adrenocorticotrophic hormones are deficient[5], and thyroxine has been shown to raise erythrocyte counts in some as yet unknown non-calorigenic fashion[6,7].

The experiments reported in the present investigation were undertaken to ascertain if exogenous thyroxine administered to hypophysectomized *Fundulus heteroclitus* could alleviate the depression of erythrocytes in the peripheral circulation, and also what effect exogenous thyroxine would produce if administered to intact fish. Three thyroxine injection experiments were performed; all fish employed in the experiments were male killifish adapted to 20° C on 8 h/day illumination. In the first two experiments, ten daily intraperitoneal injections of DL-thyroxine (Nutritional Biochemical Corporation) were administered to intact killifish adapted to fresh water, and hypophysectomized fish adapted to sea water. In the third experiment, twenty daily injections of the hormone were given to fresh water adapted intact fish. In the latter group the fish were also starved for four days in order to decrease the intrinsic activity of the thyroid gland[8]. Parallel control groups of fishes were also maintained. The dosage of thyroxine given was 5 μl/g body weight in 0·01 ml alkaline solvent. Both control and experimental fishes were autopsied after the termination of the experiments; blood was taken from the caudal artery after cutting the caudal peduncle and the total counts (per mm³ of blood) of erythrocytes, leucocytes, and thrombocytes were determined with the Unopette System.

The results are summarized in Table I. No significant change was observed in the cellular blood components in the hypophysectomized killifish in sea water after ten daily injections of thyroxine. However, the same hormonal treatment evoked a significant increase ($p < 0{\cdot}001$) in the abundance of circulating erythrocytes in fresh water adapted intact fish, with no effect on either leucocytes or thrombocytes. Chronic administration of the hormone (20 daily injections) on fishes starved for four days showed a similar significant increase in the number of erythrocytes.

TABLE I

Effects of intraperitoneal injections of DL-thyroxine, 5 μl/g body weight in 0·01 *ml alkaline solvent, on the cellular elements in the circulating blood of intact and hypophysectomized male* Fundulus heteroclitus *adapted to fresh (FW) and sea water (SW) respectively. Controls were injected with the solvent*

Experiment Group	N	Parameter M±S.E.		
		RBC × 10^6/mm^3	WBC × 10^3/mm^3	Thrombs × 10^3/mm^3
1. Control, intact FW	8	3·51±0·16	2·76±1·01	196·30±46·40
Expt'1, thyroxine (10 inj.) FW	8	4·42±0·14*	2·20±0·37	112·50±11·60
2. Control, intact FW	10	3·17±0·15	2·44±1·07	161·00±30·30
Expt'1 thyroxine (20 inj.) FW	11	3·95±0·13*	1·40±0·51	134·50±16·00
3. Control, hypophysectomized SW	6	3·05±0·38	0·83±0·29	118·30±14·80
Expt'1. hypophysectomized thyroxine (10 inj.) SW	8	2·94±0·21	1·79±1·07	111·30±9·90

* Significantly different from controls, $p < 0{\cdot}001$.

If one adheres to the mammalian concept that thyroxine administration raises red blood cell counts in the peripheral circulation of hypophysectomized animals, one would expect the hypothysectomized hypothyroid *Fundulus* to elicit a similar reaction to the hormone. Contrararily, no effect on the abundance of erythrocytes was observed in the hypophysectomized fish. The question now arises, what causes the erythrocytes to increase significantly in fresh water adapted intact fish receiving thyroxine injections ? The fact that thyroxine injection into intact fish increases the number of circulating erythrocytes but not in hypophysectomized recipients may indicate a pituitary factor being stimulated by thyroxine. In hypophysectomized fish thyroxine *per se* has no effect and the absence of the effect may reflect the absence of the pituitary factor.

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---

1. Slicher, A. M., *Bull. Bingham. Oceanogr. Coll.*, 1961, 17, 1.
2. Ball, J. N., Slicher, A. M. and Hancock, M. P., *J. Endocrinol.*, 1966, 37, xii.
3. LaRoche, B., Woodall, A. N., Johnson, C. L. and Halver, J. E., *Gen. Comp. Endocrinol.*, 1966, 6, 249.
4. Kalashnikow, G. N. and Skadowsky, S. N., *Zool. Zh.*. 1940, 19, 671.
5. Crafts, R. C. and Meineke, H. A., *Am. J. Clin. Nutrition*, 1957, 5, 453.
5. Fisher, J. W. and Crook, J. J., *Blood*, 1962, 19, 557.
7. Meineke, H. A. and Crafts, R. C., *Proc. Soc. Exper. Biol. and Med.*, 1964, 117, 520.
8. McBride, J. R., *J. Fish. Res. Board Can.*, 1967, 24, 67.

NUCLEIC ACIDS IN THE DARK AND WHITE MUSCLES OF A FRESHWATER CARP, *BARBUS STIGMA* (CUV. AND VAL.)

ALTHOGH the number of papers reporting the biochemical and physiological variations between the dark and white muscles of fishes are fairly extensive[1-6], no attempt appears to have been made to furnish a comparative account of the nucleic acid contents of the two types of muscles. The present study was undertaken to find the quantitative differences in the ribose nucleic acid (RNA) and deoxyribose nucleic acid (DNA) in the dark and white muscles of a freshwater carp, *Barbus stigma* (Cuv. and Val.).

Fishes captured by cast nets off the freshwater ponds of Aligarh (lat. 27° 34′ 30″ N, long. 78° 4′ 26″ E) were with the laboratory water at 23° ± 2° C and the dissolved oxygen concentration was maintained at 5 ± 2 ppm. At the time of investigation the specimens were killed by decapitation and the samples of the dark and white muscles were removed from the trunk region. The method for the extraction of RNA from the tissues and its determination by the orcinol reaction was the same as given by Schneider[7]. Orcinol was purified by dissolving it in boiling benzene and crystallizing it after adding hexane. A standard curve was prepared, relating the optical density to micrograms of RNA, with purified yeast ribose nucleic acid serving as the standard. DNA was extracted by the procedure of Webb and Levy[8] and estimated by Croft and Lubrans'[9] modification of Burton's[10] method of diphenylamine reaction. Diphenylamine was purified first by recrystallizing it from boiling hexane and thereafter carrying out the same treatment with petroleum ether as a substitute to hexane. The values were compared with a calibration curve, prepared by relating the color intensity to micrograms of DNA, with highly polymerized calf-thymus DNA as the standard.

For the above estimations the color intensity was read on a Bausch and Lomb Spectronic 20 Spectrophotometer.

The RNA and DNA contents of the dark and white muscles are given in Table I.

TABLE I

RNA and DNA contents of the dark and white muscles of Barbus stigma

	RNA (μg/100 mg fresh tissue) Mean ± S.E.	DNA (μg/100 mg fresh tissue) Mean ± S.E.
Dark muscle	110·491±5·237	10·286±1·028
White muscle	79·412±3·065	6·280±0·624

± S.E. = Standard error.

It is evident from the table that the RNA content of the dark muscle is significantly higher ($P < 0 \cdot 10$) than that of the white muscle. The amount of DNA contained in the dark muscle is also greater ($P < 0 \cdot 20$) than that in the white muscle. The higher level of RNA in the dark muscle in comparison to that in the white can be attributed to the greater metabolic activity in this tissue. Other biochemical and physiological investigations[11-15] also suggest that unlike the white muscle which is directly concerned with the mechanical activity, the dark muscle performs a variety of metabolic functions, resembling the liver in many respects. The apparently higher amount of DNA in the given weight of the dark muscle is due to the fact that this tissue consists of cells which are smaller than those of the white, and as a result a larger number of cells are expected to contribute to the given weight of the dark muscle, and thereby increasing the value of DNA per unit weight of the muscle sample. This has been supported by some earlier microscopical observations on the cell dimensions[16-19].

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---

1. Igarashi, H., Zama, K. and Katada, M., *Bull. Jap. Soc. Scient. Fish.*, 1957 *b*, 22, 791.
2. Nakano, T. and Tsuchiya, Y., *Bull. Jap. Soc. Scient. Fish.*, 1960, 26, 1095.
3. Buttkus, H., *J. Fish. Res. Bd. Can.*, 1963, 20, 45.
4. Ogata, T. and Mori, M., *Histochem. Cytol.*, 1963, 11, 645.
5. Mellgren, S. I. and Mathisen, J. S., *Z. Zell. forsch mikrosk. Anat.*, 1966, 71, 169.
6. Ackman, R. G. and Cormier, M. G., *J. Fish. Res. Bd. Can.*, 1967, 24, 357.
7. Schneider, W. C., In : *Methods in Enzymology*, Ed., S. P. Colowick, and N. O. Kaplan, Academic Press, New York, Vol. III, 1957.
8. Webb, J. M. and Levy, H. B., *J. Biol. Chem.*, 1955, 213, 107.
9. Croft, D. N. and Lubran, M., *Biochem. J.*, 1965, 95, 612.
10. Burton, K., *Ibid.*, 1956, 62, 315.
11. Braekkan, O. R., *Nature, Lond.*, 1956, 178, 747.
12. Mori, T., Hashimoto, Y. and Komata, Y., *Bull. Jap. Soc. Scient. Fish.*, 1956, 21, 1233.
13. Braekkan, O. R., *FisKDir. SKr. Serie Teknologiske underskelser*, 1959, 3, 42.
14. Tsuchiya, Y. and Kunü, K., *Bull. Jap. Soc. Scient. Fish.*, 1960, 26, 284.
15. Barets, A., *Archs. Anat. microsc. Morph. exp.*, 1961, 50, 91.
16. Ihurston, E. C. and MacMaster, P. P., *Food Research*, 1960, 25, 229.
17. George, J. C., *Am. Midl. Nat.*, 1962, 68, 487.
18. — and Bokdawala, F. D., *J. Anim. Morph. Physiol.*, 1964, 11, 124.
19. Nishihara, H., *Arch. histol. Jap.*, 1967, 28, 425.

CHEMICAL CONTROL OF DOWNY MILDEW OF SORGHUM

CHEMICAL control of downy mildew of *Sorghum* incited by *Sclerospora sorghi* is a much debated subject. In this article the chemical control achieved is described.

Considerable experiments were conducted using various fungicides as seed treatment and spray since 1970. Ultimately two fungicides (alkyl bisdithiocarbamates) one containing zinc (Dithane Z–78) and the other a coordination compound of zinc and manganese (Dithane M–45) were selected because of their ready availability and wide spectrum of activity. The experiment was conducted during two *Kharif* seasons, *viz.*, 1974–75 (season I) and 1975–76 (season II).

The experiments were laid out in both seasons in Random Block Design with four replications. The plot size was $4 \cdot 5$ m $\times$ $2 \cdot 7$ m. The spacing between the plots was $1 \cdot 5$ m. Spacings between rows and plants were 45 cm and 15 cm respectively. A suspectible line of *Sorghum* (DMS 652) was employed as test plant. The crop was fertilized at the rate of 15 N, 15 P, 15 K per acre. Individual plots received 400 g of the fertilizer mixture. Artificial inoculation was carried out by incorporating powdered shredded leaves of *Sorghum* containing oospores of *Sclerospora sorghi.* Disease incidence was recorded 45 days after planting.

Season I (1974–75)

1. One Kg of the seed was mixed intimately with 6 g of the active ingredient of the fungicides.
2. Seed treatment *plus* foliar spray at a concentration of 3 g active ingredient of the fungicides per litre of water.
3. Foliar spray alone.
4. Control.

The crop was sprayed first, when the seedlings just started to emerge from the soil (about 6 days after sowing). This was followed by three sprays at weekly intervals.

The data presented in Table I show that Dithane M–45 alone brought down the incidence of the disease by 63%. Seed treatment alone did not reduce the disease incidence. Foliar spray alone and seed treatment combined with foliar spray gave similar results suggesting that foliar spray is the important practice contributing to the reduction of the disease.

Season II (1975–76)

There were four spray treatments using Dithane M–45 with controls.

1. Only one spray after emergence of the seedlings,

TABLE I

Effect of Dithane M-45 and Dithane Z-78 on the incidence of downy mildew of Sorghum

	Per cent incidence of downy mildew (average of four replications)			
	Season I			Season II
	Dithane M–45	Dithane Z–78		Dithane M–45
Seed treatment alone	26·9 (31·2)	37·0 (37·4)	One spray	22·9 (28·5)
Seed treatment *plus* foliar spray	9·5 (15·1)	26·9 (31·1)	Two sprays	15·9 (23·4)
Foliar spray alone	9·6 (16·2)	27·1 (30·7)	Three sprays	5·7 (13·4)
Control	25·6 (30·1)	31·5 (33·9)	Four sprays	2·8 (9·0)
			Control	17·6 (21·2)
	S.E. = 5·33 C.D. at 5% = 12·04	S.E. = 5·16 Not significant		S.E. = 3·50 C.D. at 5% = 7·6 C.D. at 1% = 10·6

(Figures in the parentheses show transformed values)

2. Two sprays, first after emergence and the second a week after.
3. Three sprays, first after emergence the second and the third at weekly intervals.
4. Four sprays, first after emergence and second, third and fourth at weekly intervals.

Data presented in Table I show that the disease can be controlled to the tune of 84% by a four spray schedule. The results achieved through a three spray schedule did not differ significantly from a four spray schedule. The disease incidence in control plots and single spray treatments did not differ significantly. It becomes quite obvious that the downy mildew of *Sorghum* can be controlled quite effectively by a spray schedule using zinc manganese ethylene bis dithiocarbamate (Dithane M–45).

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SUSCEPTIBILITY OF RAMIE WITH DIFFERENT GUM CONTENTS TO MICROBIAL DAMAGE

RAMIE, a bast fibre obtained from the plant *Boehmeria nivea,* is well known for its strength, fineness and durability. Raw ramie fibre obtained by decortication contains a lot (10–40%) of gummy matter, carbohydrate in nature. In order to make ramie usuable, it is first degummed to a certain extent with alkali, washed and then dried. It has been observed in this laboratory that ramie fibres of different gum contents are not equally suspectible to microbial damage when kept in storage. In order to assess the suitability of ramie products for different uses, a study was made to find out the comparative susceptibility of remie fibres with different gum contents to microbial damage.

Decorticated ramie for this study was obtained from Ramie Research Station, Sorbhog, Assam (variety R 1452, 2nd cutting, 1971) and degummed chemically to different gum contents[1,2]. To maintain uniformity of the samples, the middle portions of the reeds were taken, cleaned and sampled under identical conditions. Bundles of each sample, 8" in length and 15 g in weight were incubated at about 30° C, moistened with 200% distilled water in sterile petri dishes under covered condition for different periods of time. Loss of moisture in the petri dishes during incubation

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TABLE I

Bundle tenacity of ramie with different gum contents after incubation*

Gum Content %	Bundle strength (g/tex) after incubation with 200% moisture for						Bundle strength (g/tex) after exposure for 90 days to normal atmospheric conditions
	0 day	15 days	30 days	45 days	60 days	90 days	
23·0	29·1	10·0	1·9	1·9	0**	0	20·8
20·3	28·3	5·2	0	0	0	0	24·0
14·8	30·4	14·5	0	0	0	0	19·8***
8·2	33·3	15·5	10·7	6·8	6·8	3·0	23·4
6·9	34·4	24·3	22·3	14·1	12·7	13·5	24·5
3·0	33·3	27·0	26·8	26·3	24·3	21·1	26·6

* Results expressed as average of 3 replications.

** Bundle strength 0 g/tex indicates that the fibre sample was so weak that it could not be tested by the method described.

*** This anomalous value might be due to sample variation.

period was made up aseptically with distilled water once a week. Under the above conditions, only the microorganisms present in the fibres themselves were allowed to act on the fibres. At definite intervals of time, representative samples were taken out, washed first with water and then with rectified spirit and finally dried in air. This method facilitated quick drying of the samples. The samples were then tested for bundle strength in J.T.R.L. bundle strength tester[3] in order to assess the extent of retention of fibre strength. A set of samples was simultaneously kept exposed to atmosphere in the room under normal conditions without addition of any extra moisture for 90 days and was tested for bundle strength.

Results are presented in Table I which shows that more the gum content of the fibre, more is the extent of damage. On visual observation, it was also found that the extent of fungal growth was more or less proportional to the gum content.

α-cellulose[4] of ramie is highly crystalline and as such microorganisms cannot grow directly on it. Gum of the ramie fibre being a heterogeneous carbohydrate complex serves as a ready nutrient and source of energy to the damaging microbes. As a result, the fibre with more gum becomes more susceptible to microbial damage, but ramie with about 6–7% gum is fairly resistant to damage (Table I) compared to those of higher gum content. Degumming to this extent can be conveniently and economically achieved in industry.

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---

1. Mazumder, M. C., Sen, S. K. and Dasgupta, P. C., *Indian Textile Journal,* 1975, 85, 135.
2. —, — and —, *Ibid.* (In press).
3. Bandyopadhyay, S. B., *Jute Bulletin,* 1965, 27, 278.
4. Luniac, B., *Textile Quarterly,* 1954, 4, 161.

THE EFFECT OF A DESYNAPTIC GENE ON B-CHROMOSOMES IN PEARL MILLET

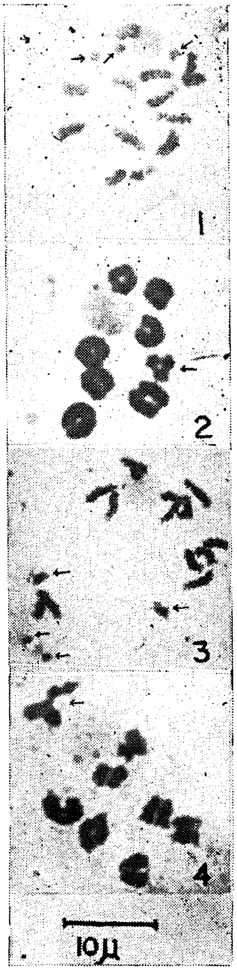
THE morphology of B-chromosomes at pachytene and their pairing behaviour at diakinesis and metaphase I in *Pennisetum typhoides* S and H were reported earlier (Pantulu[1]; and Pantulu and Manga[2]). B-chromosomes, up to 3 had no significant effect on the mean chiasma frequency of A-chromosomes but increased the variance of the mean. However, the presence of 5 or more B's produced deleterius effect like desynapsis of A-chromosomes. In *Pyrgomorpha krausi* (Lewis and John[3]) and in rye (Jones and Rees[4]) the behaviour of B's was suggested to be influenced by the A-chromosome genotype and the effect of B's on A-chromosomes was considered to result from an interaction between the two types of chromosomes.

The present investigation was aimed at a study of the effect of a desynaptic gene, which produced desynapsis of the A-chromosomes, on the pairing of B's in *P. typhoides.* Among the progeny of a plant treated with 0·4% colchicine one plant was a desynaptic with 14 univalents in a large number of cells and the mean chiasma frequency in this plant was 0·68 per cell. The desynaptic condition was inherited as a monogenic recessive. In F_2 and further generations desynapsis was variable with mean chiasma frequencies ranging from 2·91 to 6·93 per cell. Plants showing extreme desynapsis were totally sterile on selfing and therefore were maintained by controlled pollination using desynaptic plants as females. Even after controlled pollination seed set was about 10%.

The lines carrying B-chromosomes were maintained by selfing for over 10 generations. The B's when originally recorded had subterminal centromere with a heterochromatic short arm. In the long arm a heterochromatic block adjacent to the centromere and 8 chromomeres in the euchromatic long arm were present. The number of B's varied within and between plants from 1 to 4, but there were no detectable differences either in length or morphology between the B's. But in plants after several generations of selfing in addition to B's with the above morphology, designated as standard type, two other types were present with different sizes and morphology. These are designated (1) deficient B's in which 2–3 chromomeres in the long arm were missing and (2) iso-B's, derived from the long arm of the deficient B's.

Deficient and iso-B's were also different in their effects on A-chromosome chiasma frequency. In non-desynaptic diploid plants 2–4 deficient B's produced a decrease in the mean A-chromosome chiasma frequency while 1–2 iso-B's produced an increase in the mean.

In a cross between a desynaptic plant without B's and a non-desynaptic plant with 2 standard and 2 deficient B's, four F_1 plants were with B-chromosomes. One of the plants had one standard and two deficient B's, while the other 3 carried two standard and two deficient B's. As expected all the F_1 plants were normal (non-desynaptic), and there was no detectable difference in the mean chiasma frequency of the A-chromosomes as compared to normal plants without B's. Due to unfavourable weather conditions F_2 progency could be obtained from one F_1 plant only. There were 17 plants of which 13 were non-desynaptic and 4 were desynaptic thus segregating in the expected 3 : 1 ratio. Of the 4 desynaptic plants, one was without B's, one plant had three standard B's and two plants had two standrad and two deficient B's each. Among the 13 normal plants, two were without B's, 3 had only standrad B's and 8 plants had standard and deficient B's in different proportions.



FIGS. 1–4. Fig. 1. Diakinesis in desynaptic plant showing 3 standard B's ; 5 A-chromosome bivalents and 4 univalents. Fig. 2. Diakinesis in normal plant showing trivalent association of 3 standard B's and 7 A-chromosome bivalents. Fig. 3. Diakinesis in desynaptic plant showing 2 standard *plus* 2 deficient B's and 5 A-chromosome bivalents and 4 univalents. Fig. 4. Diakinesis in normal plant showing quadrivalent associations of 2 standard *plus* 2 deficient B's ; 7 A-chromosome bivalent.

For the study of the effect of desynaptic gene, on the pairing of B-chromosomes, meiosis in the PMCs of desynaptic plants with B's was compared with that in non-desynaptic plants with the corresponding number and type of B's. When standard

TABLE I

Associations of A and B-chromosomes at Diakinesis in non-desynaptic and desynaptic plants without and with B's

Plant	A-chromosome associations			B-chromosome associations				Total no. of cells
	Ring	Rod	Uni	Qua	Tri	Bi	Uni	
Desynaptic without B's	145 (1·45)	300 (3·00)	519 (5·10)	..	..	..	..	100
Desynaptic with 3 St. B's	210 (2·10)	318 (3·18)	344 (3·44)	..	..	3 (0·03)	295 (2·95)	100
Desynaptic with 2 St. + 2De.B's	233 (1·16)	488 (2·44)	1358 (6·79)	..	..	47 (0·235)	360 (3·3)	200
Non-desynaptic without B's	1116 (5·58)	284 (1·42)	..	..	..	..	..	200
Non-desynaptic with 3 St. B's	590 (5·90)	110 (1·10)	..	..	73 (0·73)	26 (0·26)	29 (0·29)	100
Non-desynaptic with 2 St. + 2 DeB's	1180 (5·90)	220 (1·10)	..	81 (0·405)	67 (0·335)	42 (0·21)	42 (0·225)	200

'( )'—Numbers within brackets indicate the mean number of associations per cell.

St—Standard B's. De—Deficient B's.

B's alone or when standard and deficient B's were present together in the desynaptic plant the B's did not form multivalents and the univalent frequency was much higher than that in the normal plants of corresponding B classes (Table I; Figs. 1 and 2). The absence of multivalent formation and the very low bivalent frequency in the desynaptic plants were the result of low B-chromosome chiasma frequencies. The mean B-chromosome chiasma frequencies in the desynaptic plants were significantly lower than those in the normal plants of corresponding B-classes. When three standard B's were present in the desynaptic plants their mean chiasma frequency per cell was 0·031 whereas in the normal plants it was 2·12. Similarly, in desynaptic plants with two standard and two deficient B's the mean chiasma frequency per cell was 0·162 and in normal plants of corresponding B class the value was 3·01. These results clearly show that the effect of the desynaptic gene on the association and chiasma freqnency of B-chromosomes is similar to that on the A-chromosomes.

The effect of B chromosomes on A-chromosome associations was studied taking the desynaptic plant without B's as the control. When three standard B's were present in the desynaptic plant there was a significant increase in the A-chromosome bivalent frequency and decrease in univalent frequency. In plants with standard *plus* deficient B's there was a significant increase in the univalent frequency of the A-chromosomes. In normal plants with three standard B's or two standard *plus* two deficient B's the B's had no effect on the A-chromosome associations or chiasma frequencies.

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1. Pantulu, J. V., *Curr. Sci.*, 1960, 29, 28.
2. —, and Manga, V., *Genetica,* 1975, 45, 237.
3. Lewis, K. and John, B., *Chromosoma,* 1959, 10, 589.
4. Jones, R. N. and Rees, H., *Heridity,* 1967, 22, 333.

ROLE OF COMPONENT COMPENSATION IN IMPARTING GENETIC HOMEOSTASIS FOR FRUIT PRODUCTION IN AUTOGAMOUS TOMATO CULTIVARS

RECENTLY, a number of theories have been advanced to explain the phenomenon of genetic homeostasis in heterozygote population[1,3]. However, the precise information about the mechanics of homeostatic mechanism in stabilising the overall fruit production in homozygotes and particularly in tomato genotypes is rather inadequate. Such a mechanism in tomato could be attributed to the component compensation and the mutual balance of a few or a number of characters with different rates of changes in building up overall fruit production. It may be likely that in stable cultivars, the characters influencing the overall fruit production might be compensating in such a way that the stability of total fruit production is not lost but is maintained regardless of the fluctuations in the environmental parameters. In the present work, this hypothesis was basically put to test and a detailed genetic analysis was conducted with 25 cream cultivars combed out from a tomato germplasm of 228 varieties maintained at this University. They were grown under four different environment conditions (Winter, 1972 ; Summer and Winter, 1975 and Summer, 1974) in a randomised block design with three replications. Observations were recorded on fruit yield and seven characters known to be influencing fruit production in tomato.

As a first step towards identifying marker characters whose stability leads to the homeostasis in the total fruit production, correlation coefficients were worked out between the various stability parameters (bi's) among the yield and the components of the yield. The results obtained have been given in Table I. Stability parameters were estimated as suggested by Eberhart and Russel (1966)[2]. Stability of fruit yield has been related to stability of height of plant, days to first fruit set, and number of inflorescences per plant.

TABLE I

Direct and indirect effects of the component characters to induce genetic homeostasis in tomato

Sl. No.	Characters	1	2	3	4	5	6	7	Correlation with genetic homeostas is of fruit production
1.	No. of days to first fruit set	0·419	0·034	0·020	−0·126	−0·107	−0·006	−0·014	0·221
2.	No. of days to first fruit ripening	−0·185	−0·077	0·019	0·184	0·104	−0·017	0·039	0·068
3.	Height of plant (cm)	0·021	−0·004	0·401	0·264	−0·086	−0·067	−0·041	0·488*
4.	No. of primary branches per plant	0·095	0·026	−0·191	−0·554	0·113	−0·030	−0·00	−0·582**
5.	No. of inflorescences per plant	0·109	0·019	0·084	0·152	−0·410	0·184	0·022	0·159
6.	No. of fruits per plant	0·006	−0·003	0·069	−0·043	0·196	−0·386	0·023	−0·185
7.	No. of locules per fruit	0·041	0·021	0·114	−0·076	0·061	−0·061	−0·146	−0·047

*, ** Significant at 5% and 1% level of probability respectively.

The underlined figures indicate the direct effect, the remaining ones (1 to 7) are the indirect effects.

The detailed statistical analysis (Table I) revealed that the stability of a number of primary branches had a negative influence on the stability mechanism, its direct effect on the stability of the fruit yield being − 0·554. Days to fruit set and height of plant had the maximum direct effect (0·419 and 0·401 respectively) on the stability of the yield expression. The tomato cultivars, namely, Sanmarsano, Roma and 2152–14 were observed to be most stable and possess a balanced homeostatic genetic system. Invariably, these varieties were highly stable for the characters days to first fruit set and height of the plant. These were, however, highly unstable for the character, number of primary branches. Varieties S_5 First, Marglobe, HS101, Momor and Best Of All were observed to be highly unstable varieties. In these cultivars, days to first fruit set and height of plant were highly unstable. However, these were stable for the character, primary branches per plant. From these observations, it could be deduced that in a homeostatic genotype of tomato, the component characters like days to first fruit set, plant height and number of primary branches act in a compensating manner with each other in the changing environments and provide the tomato cultivar the ability to give a buffering mechanism and the consistent performance. On the other hand, in the unstable varieties, the above-mentioned characters do not compensate each other to adapt them to the changed environmental parameters and hence the cultivars exhibit a lower expression of fruit production under stress conditions.

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---

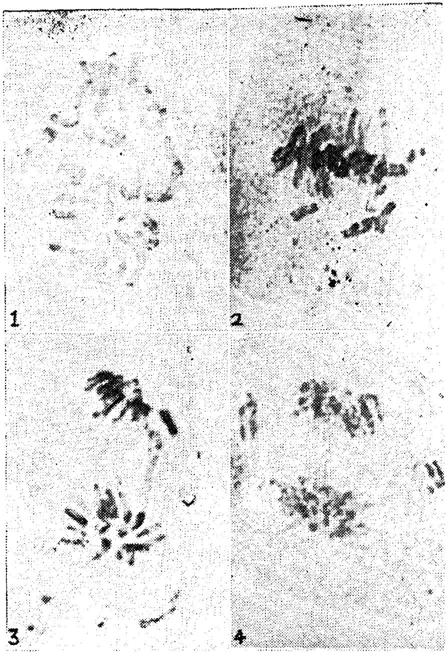
1. Bains, K. S. and Gupta, V. P., *Indian J. Genet.*, 1972, 32, 306.
2. Eberhart, S. A. and Russell, W. A., *Crop Sci.*, 1966, 6, 36.
3. Grafius, J. E., *Agron. J.*, 1956, 48, 419.

EFFECT OF GAMMA-RADIATION ON MITOSIS

GAMMA-RADIATION has now been widely used for the artificial productios of genetic variants in the plants. In the present study, the effect of gamma-irradiation on mitosis of barley (*Hordeum vulgare* L.) root tips was investigated.

Seeds of cultivated six rowed barley variety, K_{12}, were irradiated with four doses of gamma-rays from Co^{60} source at Bhabha Atomic Research Ctntre, Trombay, Bombay. The different doses were 20 Kr, 30 Kr, 40 Kr and 50 Kr. The irradiated seed samples were germinated on different moist blotting papers and when the root tips attained a height of 1 to 1·5 cm., they were collected and fixed in 1 : 3 acetic-alcohol. After 24 hr interval, these root tips were transferred in permanent fixative of 70% alcohol and kept in a refrigerator. Before squashing, the root tips were hydrolysed in 1 N HCl at 60° C for 5 minutes and then squashed with 2 or 3 drops of aceto-orcein stain on clean slides under microscope. Observations were recorded on the number of dividing cells at different stages of mitosis and the various types of abnormalities produced by gamma-radiation are recorded in Table I.

Except with the 30 Kr dose, increase in radiation dose increases the number of cells in prophase stage and the relationship between radiation doses and cells in prophase stage was found linear. It can, therefore, be concluded that the radiation induces inhibition of mitosis. 'Relationship between the doses of radiation and cells in metaphase, anaphase and telophase was, however, not linear, and highest percentage of metaphase, anaphase and telophase cells were recorded at 30, 40 and 50 Kr doses respectively.



FIGS. 1–4. Fig. 1. Showing chromosomes with fragments. Fig. 2. Chromosomes clumping at metaphase with fragments. Fig. 3. A dicentric chromosome bridge at anaphase. Fig. 4. Dicentric chromosome bridge with lagging chromosomes at anaphase.

Tht different types of mitotic aberrations observed in the present study were : chromosome breaks (fragments), bridges, stickiness and micro-nuclei. Chromosome fragments were found in all the four doses of radiation and the highest frequency was recorded at 50 Kr followed by 40 Kr, 30 Kr and 20 Kr. The relationship between the frequency of radiation doses and chromosome fragments was found linear. This type of linear relationship between the frequencies of the two types of chromosomal aberrations (fragments and bridges) and X-ray doses was reported earlier in many plant materials by Sax and Brunfield (1943), Caldecoll *et al.* (1959) and Yagy and Morris

TABLE I

Frequency of dividing cells and type of chromosomal aberrations at different mitotic stages in irradiated barley (Hordeum vulgare) *material*

Radiation Dose	Total No. of cells studied	Dividing cells at different mitotic stages			
		Prophase (%)	Metaphase (%)	Anaphase (%)	Telophase (%)
20 Kr	400	87·0	8·0	4·0	1·0
30 Kr	500	86·0	9·0	3·0	1·0
40 Kr	300	89·0	6·0	4·0	1·0
50 Kr	300	89·0	5·0	3·0	2·0

Abnormal cells observed		Types of abnormalities							
No.	Percentage (%)	Fragments at metaphase		Bridges at anaphase		Stickiness at metaphase		Micronuclei at telophase	
		No.	%	No.	%	No.	%	No.	%
4	1·0	2	0·5	2	0·5	..	..	..	..
11	2·0	5	1·0	4	0·8	1	0·2	1	0·2
12	4·0	4	1·3	5	1·7	1	0·3	2	0·7
18	6·0	5	1·7	7	2·3	2	0·7	4	1·3

(1957). Chromosome bridges were, however, found in anaphase and telophase stages and the highest frequency was observed at 50 Kr dose. Most of the chromosome bridges observed were double stranded and criss-crossed. The presence of paired dicentric chromosome bridges and fragments suggest that the resting chromosomes behave as monopartite and the chromosome bridges and fragments are the result of breakage at chromosome level rather than chromatid level. Also the frequency of fragments per cell was more than that of the chromosome bridges, indicating the chromosomes deletion in addition to the formation of dicentric chromosomes.

As reported by Marquard (1938) and Darlington (1945), chromosome stickiness in the result of the alteration of surface properties of the chromosomes due to radiation. In the present investigation, chromosome stickiness was observed in both metaphase and anaphase stages and did not show any relationship with the radiation doses. Hence it can be concluded that stickiness does not prevent anaphase movement, except some hindrance of chromatid separation.

High frequency of occurrence of micronuclei was found in cells when the percentage of fragments was high. This indicates that some of the fragments would form micronuclei and the relationship between the doses of radiation and frequency of micronuclei was linear. Also, it could be concluded that the occurrence of micronuclei is an efficient measure of chromosome damage.

The study was carried out under the guidance of Dr. K. Das, Head of the Department.

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---

1. Catcheside, D. G., *Advances in Genetics*, 1948, 2, 271.
2. Caldecott, R. S., *Radiation Res.*, 1955, 2, 239.
3. Jain, H. K. and Majumdar, *Cur. Sci.*, 1959, 28, 8.

PROMISING NEW MULTIPLE INSECT RESISTANT RICE VARIETIES

THE gall midge, the stem borers and the vectors of rice virus disease, *i.e.,* leaf hoppers and the planthoppers constitute the major in insect pests of rice that cause considerable yield losses annually in the different parts of the country. Effective and economic chemical control of these is rather difficult to achieve, as it is dependent on factors like timing, dosage, method and frequency of application of insecticides. Thus evolving high yielding rice varieties combining field resistance/tolerance to the major pests would provide an effective alternative.

With the objective of developing lines possessing resistance/tolerance to pests, varieties in the genetic stock collection being maintained at the Central Rice Research Institute, Cuttack, are regularly screened for their reaction to pests to provide donors for utilization in breeding programmes. This had resulted in the identification of a number of resistant donors for the different insects some of which are listed below.

Donors	Varieties
Gall midge ..	Ptb 10, Ptb 18, Ptb 21, Leaung 152, ARC 6158, ARC 5984, ARC 6221, JBS 446, JBS 673, MNP 30, MNP 471.
Stem borer ..	TKM 6, Ptb 18, Ptb 21, Leaung 152, CB I, CB II, Topa I, W 1263, GEB 24.
Leaf hopper ..	Ptb 18, Ptb 21, TKM 6, W 1263, Ptb 2, Ptb 7
Planthopper ..	TKM 6, ARC 6238, AC 255, Ptb 21, Ptb 33

The degree of resistance of these donors is later confirmed under artificial conditions. The varieties exhibiting a desired level of tolerance/resistance to one or more major insects are crossed to selected high yielding types to develop high yielding strains combining good yield potential and desired level of resistance.

Forty cultures combining good yield potential and a desired level of resistance to more than one insect pests, were subsequently identified from seven crosses involving four donors. From these cultures, based on data collected from different sources, four were finally retained. The information on these cultures along with that of Jaya is presented in Table I. These cultures mark a significant improvement over the existing varieties. In addition to the data provided in Table I, it may be said that CR 94-MR-1550 is suitable for late planting in *kharif* (*e.g.,* after the harvest of jute) and can tolerate transplanting of aged seedlings (upto 60 days age) without any appreciable reduction in yield. Even under 0 nitrogen level, this had yielded better than several released high yielding varieties.

CR 139–1047 has been reported to possess a high degree of resistance to bacterial leaf blight in international baterial blight nursery trials conducted at many test locations in India, Ceylon, Thailand, Indonesia, Philippines, etc.

The new dwarf rices released thus far in India possess low level of field resistance to common rice pests and as a result, the varieties have already become susceptible to major pests in several parts of the country. In fact, in parts of Kerala, the incidence of planthoppers has been reported to have

TABLE I

Insect and disease reaction and yield of promising multiple insect resistant cultures

Culture	Cross	Reaction to							Duration (days)	Yield (t/ha) without prot.	Grain character
		Gall midge	Stem borer	Leaf hopper	Plant-hopper	RTV	BLB	Blast		*Kharif*	
CR 94-MR-1550	(Ptb 21 × Ptb 18) IR 8	R	MR	R	MR	R	MR	S	130	3·5	Medium bold red rice
CR 57-MR-1523	IR 8 × Ptb 21	R	MR	R	R	R	S	MR	124	3·2	Medium bold white rice
CR 138–928	Jaya × TKM 6	S	R	R	MR	MR	MR	MS	137	3·0	Long slender white rice
CR 139–1047	(TKM 6 × IR 8) TKM 6	S	R	MR	MS	MS	R	R	135	3·0	Long slender white rice
Jaya (susceptible check)	T (N) 1 × T 141	S	S	S	S	S	S	MS	140	1·5	Long bold white rice

reached an alarming situation. The large scale introduction of the multiple resistant varieties in the different insect problem areas would reduce the losses in yield and directly contribute to increased production. Minimum plant protection especially in the seedling stage might be taken as an additional precaution in pest endemic areas.

The authors are grateful to Dr. S. Y. Padmanabhan, Director, Central Rice Research Institute, Cuttack, and Dr. R. Seetharaman, Project Director, All India Coordinated Rice Improvement Project, Hyderabad (formerly, Head, Division of Genetics, CRRI, Cuttack), for their valuable suggestions and encouragement.

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FIRM SEEDS IN THE FINGER MILLET (*ELEUCINE CORACANA*, G.)

There is very little published data concerning dormancy in finger millet (ragi). The present work deals with the dormancy in ragi and the methods of overcoming the same.

A sample of 'Purna ragi' was subjected to standard germination test as per *Seed Testing Manual* (1969). Due to the presence of large number of fresh ungerminated seeds, dormancy was suspected to prevail in the seeds. The fresh ungerminated seeds were kept in the germination paper with regular supply of adequate moisture upto 45 days and tested for viability by pricking the seeds with a fine needle at an interval of one week as tried by Delouche (1964) on rice. Suspecting the prevalence of dormancy in the seeds, fresh samples were drawn from the original lot and tested for germination after giving the treatments as in Table I.

Results and Discussion

The germination percentages obtained after the end of test are as given in Table I.

TABLE I

Showing germination percentages

Treatments	RI	RII	RIII	RIV	Total	Mean
1. 0·2% KNO_3	95	95	97	94	381	95
2. Soaking overnight	86	88	90	89	353	88
3. Soaking for 3 days	92	89	92	90	363	91
4. Scarification	85	82	80	84	331	83
5. Sand as substrata	38	40	40	35	153	30
6. Control (paper)	25	28	29	26	108	27

C.D. = at 1% level 3·9157. S. Em. = 1·879.

The statistical analysis showed that the results are highly significant. From the data (Table. I) it is evident that germination of ragi seeds was improved by various seed treatments. Methods like treatment with 0·20% solution of potassium nitrate and soaking were found more effective than scarification. It is probable that efficient scarification may bring up the level of germination to that of other methods. Delouche (1966) while working with graminae seeds reported that impermeability of the pericarp-seed coat complex to oxygen (but not to water) was the main cause for the dormancy which could be overcome by rupturing the pericarp-seed coat complex mechanically. In the present investigation the treatments have helped in softening or by mechanical breaking the seed coat complex, which, probably acted as a physical barrier for the entry of oxygen into the seeds. The ungerminated seeds when pricked with fine needle germinated well. Such results were also obtained by Delouche *et al.* (1964) on rice, Bass (1955) on wheatgrass seeds, Laude (1949) on oatgrass and Wayne *et al.* (1962) on sorghum. Among the methods used for breaking the dormancy, scarification appears to be fairly simple. The other methods are also quite efficient.

Agricultural Department (Seed Testing), Hebbal, Bangalore, *October* 6, 1975. — G. N. Kulkarni. V. Basavaraju.

---

1. Bass, L. N., *Proc. Assoc. off Seed Anal,* 1955, 45, 102.
2. Challam, G. V., Amir Singh, and Doughlas, J. E., *Seed Testing Manual,* 1969.
3. Delouche, J. C. and Nguyen, N. T., *Proc. Assoc. off Seed Anal.,* 1964, p. 5441.
4. Delouche, J. C., *Proceedings Seed Improvement Training Courses,* 1966, 1, 179.
5. Laude, H. M., *Jour. Amer. Soc. Agron.,* 1949, p. 401.
6. Wayne, G., Wright and Kinch, L. R. C., *Proc. Assoc. off Seed Anal.,* 1962, 52, 169.

EFFECT OF HYDROGEN-ION CONCENTRATION ON THE ACTIVITY OF INCLUSION BODIES OF *SPODOPTERA LITURA* NUCLEAR POLYHEDROSIS VIRUS

THE subtle influence of pH on the activity and dissolution of polyhedral inclusion bodies (PIB) of nuclear polyhedrosis virus (NPV) of *Hellothis zea* and *Trichoplusia ni* has been reported earlier[1,2]. The present communication deals with the effect of different levels of pH on the activity of PIB of *Spodoptera litura* F.

The purified suspension of PIB of *S. litura* NPV obtained by differential centrifugation was brought to different pH levels[3], *viz.*, 9·8, 8·4, 7·0, 5·6 and 3·8 by mixing with appropriate buffers. Virus with a concentration of $4 \cdot 8 \times 10^6$ PIB/ml in the respective buffers was incubated at room temperature (28° C) for one day and fed to twenty early fourth instar larvae along with host leaf. Larvae fed with virus free leaf dipped in phosphate buffer, pH 7·0 served as control and mortality counts were recorded at regular intervals.

The study revealed that at neutral pH 7·0 highest larval mortality (100%) was recorded within 4·55 days. At pH 8·4 and 5·6, despite a general decrease in the larval mortality (60 and 50%); the activity remained unaffected. Three to four fold reduction in the activity of virus was obtained at pH 9·8 and 3·8 which concomitantly prolonged the mortality time, through 8·66 to 9-0 days with 30 and 20% mortality respectively. Admittedly cent per cent pupation was recorded in control.

The present results add additional support to the views of Bergold[4] who stated that the pH of the suspending media should not drop below 5·0 or rise above 8·5 in order to prevent the dissolution of PIB of NPV of different insects. Falcon[5] suggested that basic pH of the cotton leaf surface might inactivate the NPV of *Heliothis* virus, in the present study it was observed that even though the tissue pH of castor leaf was 5·9, it did not affect the activity of the virus when the latter was brought to pH 7·0. It evidently shows that the tissue pH does not materially alter the activity of the virus. The results presented here clearly indicate that the virus should be maintained at near neutral pH for the effective control of insects.

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1. Ignoffo, C. M. and Garcia, C., *J. Invertebr. pathol.*, 1966, **8**, 426.
2. Gudauskas, R. T. and Canerday, T. D., *Ibid.*, 1968, **12**, 405.
3. Dawson, R. M. C., Daphne, C. Elliot, Elliot W. H. and Jones, K. M., *Data for Biochemical Research*, Oxford University Press, London, 1969, p. 269.
4. Bergold, G. H., " Viruses of Insects " In: *Handbuch der virus forschung.* (C. Hallauer and K. F. Meyer, ed.), Springer Vienna, 1958, **4**, 60.
5. Falcon, L. A., "Microbial control as a tool in integrated control programmes," In: *Biological Control* (C. B. Huffaker, eds.), Plenum Press, New York, 1971, p. 511.

ROLE OF *LEPTOCORISA ACUTA* THUN. IN THE SPREAD OF BACTERIAL BLIGHT OF RICE

THE source of inoculum for the secondary spread of bacterial blight of rice in the field consists mainly of the bacterial cells released through the guttation drops or in the coze from leaves injured due to strong typhoonic rains (Anonymous, 1971); and insects are rarely involved. However, studies on the role of insects in the spread of bacterial blight of rice were undertaken following the observation that in many cases blight symptoms were initiated from the punctures on leaves which resembled insect feeding sites. The pathogen, *Xanthomonas oryzae* could easily be isolated from such lesions, and the severity of the disease coincided with high populations of small insects and *Leptocorisa acuta* Thun.

The insects were collected daily early in the morning and late in the evening and were separated into the respective types, crushed and inoculated on T (N)-1 plants separately. Plants inoculated with distilled water served as control. Blight symptoms developed after 2 weeks only in plants inoculated with crushings of *Leptocorisa.* No symptoms were seen in the other treatments (Table I). Once the role of *Leptocorisa* was demonstrated, the experiments were elaborated further to determine whether the insects carried the inoculum externally or internally. Bugs were, therefore, collected and divided into two lots. One lot was surface-sterilized with mercuric chloride (0·1% for 5 min.). The sterilized and unsterilized bugs were crushed separately and the supernatants inoculated on T (N)-1 plants. The

TABLE I

Pathogenicity tests with insect macerates

Test material	No. of leaves Inoculated	No. of leaves Infected	No. of leaves % infection	Lesion length* (mm)
Heterogeneous population of insects (including *Leptocorisa*)	25	13	52·0	103
All other insects (excluding *Leptocorisa*)	35	0	0	0
Leptocorisa (without surface sterilization)	38	20	52·0	141
Leptocorisa (after surface sterilization)	35	0	0	0
Control (water)	35	0	0	0

* Average length of rice leaf: 210 mm.

temperature ranged between 26 and 38° C and relative humidity was 95% throughout the experiment. After 2 weeks the disease was noticed only in the plants inoculated with the crushings from unsterilized bugs, thereby clearly indicating that the pathogen was present on the surface of the insect.

The mechanical transmission of bacterial blight of rice by *Leptocorisa* resulting in the secondary spread of the disease has been established for the first time. Chewing insects and also sucking insects like *Nephotettix* sp. have been reported to spread the disease in the field and the chewed areas were found to serve as points of entry for the pathogen (AICRIP, 1968).

Under Indian conditions, the *Leptocorisa* population is known to increase only when there are early summer showers (relative humidity 80–82%) with an optimum temperature of 27° to 28°.C (Reddy, 1968). Therefore, effective insecticidal control measures should be applied to keep down the pest population and thus reduce the incidence of the pest as well as the disease.

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---

1. Anonymous, *Progress Report, All India Coordinated Rice Improvement Project,* 1968, 4·1–4·22 (Mimeographic).
2. —, *Ibid.*, 1971, 1, 4·1–4·29 (Mimeographic).
3. Reddy, D. B., "Pests of rice." In : *Plant Protection in India,* Allied Publishers, Calcutta, 1968, p. 89.

EFFECT OF A MORPHACTIN ON GROWTH AND FLORAL MORPHOGENESIS IN *NIGELLA DAMASCENA*

MORPHACTINS (derivatives of fluorine-9-carboxylic acid) affect various physiological and morphogenetic processes of a variety of plants[1,2]. Varied effects on flower development have also been reported[3-6]. The present communication deals with the effect of a morphactin (EMD-7311 W) on growth and floral morphogenesis of *Nigella damascena* L.

Eight-week-old plants of *N. damascena* were sprayed with 10, 25, 50, 100, 250 and 500 ppm of EMD–7311 W. Triton X-114 at 0·1% was used as surfactant. For controls, one set was treated with the surfactant only, while the other set remained untreated. Ten plants were used for each treatment.

All the concentrations used affected growth, and development. The treated plants, generally, showed reduced activity of shoot apical meristem. Within a week after treatment, the leaves of plants treated with 100, 250 and 500 ppm rolled inward, and some branches started producing almost entire leaves instead of deeply-pinnatifid leaves. Four weeks after treatment when, presumably, the effect of chemical no longer persisted these branches bore normal leaves.

In the plants treated with 250 and 500 ppm of morphactin, the visible appearance of floral bud was greatly delayed. None of the concentration used promoted flower formation.

Several abnormalities were observed in the flowers of treated plants. In a few cases leafy bracts fused with floral buds, and such buds never anthesized. In some flowers the perianth became highly dissected or, at times, gamophyllous.

Reduction in the number of stamens occurred in treated plants (Table I).

Some stamens also became transformed into carpellate structures, and 5–7 additional ovaries developed in a flower. Occasionally, the carpels fused basally but were free at the apex exposing the ovules (Fig. 1). The production of twin ovaries, numerous ovaries of different size, enlarged ovary, increase/decrease in the number of styles, and fusion of styles were frequent.

TABLE I

Effect of morphactin on the number of stamens

Flowers* %	Concentration of morphactin ppm				
	0	10–50	100	250	500
With reduced number of stamens	0	0	40	45	48
Stamen-less	0	0	20	48	52

* Average of 50 flowers..

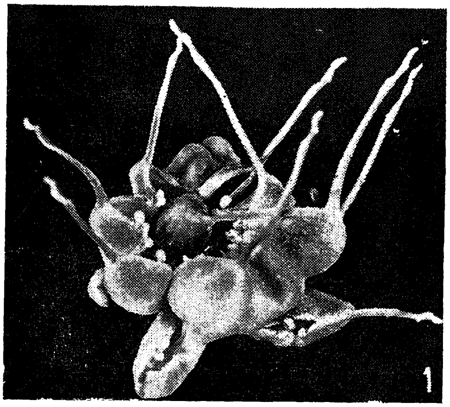

FIG. 1. Carpels showing exposed ovules, × 2·5.

These observations indicate that morphactin suppresses the stamens in *N. damascena* rendering hermophrodite flowers female. The selective male sterility induced by morphactin suggests a possible application in breeding procedures. The possibility of enhanced seed-yield due to formation of extra carpels may be useful in hybrid seed production. Further work to elucidate the mechanism of action of morphactin is in progress.

I am grateful to Dr. S. S. Bhojwani and Professor B. M. Johri for valuable advice, and to Dr. G. Schneider of E. Merck AG, Darmstadt, for a gift of morphactin (EMD-7311 W). The award of Junior Research Fellowship by C.S.I.R., New Delhi, is gratefully acknowledged.

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1. Schneider, G., *Ann. Rev. Plant Physiol.*, 1970, 21, 499.
2. —, *In* H. Kaldwey and Y. Vardai (eds.), "Hormonal Regulation in Plant Growth and Development," *Proc. Adv. Study Inst. Izmir (Turkey)*, 1971, Verlag Chemie, Weinheim, 1972, p. 317.
3. Sankhla, N., *Zeit. Pflanzenphysiol.*, 1969, 61, 350.
4. Mohan Ram, H. Y. and Jaiswal, V. S., *Naturwissenschaften*, 1971, 58, 149.
5. Uma, M. C., *Curr. Sci.*, 1972, 41, 114.
6. Jayakar, M., *Ibid.*, 1972, 41, 849.

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REVIEWS AND NOTICES OF BOOKS

Integral Equation Via Imbedding Methods. By H. H. Kagiwada and R. Kalaba, Applied Mathematics and Computation, No. 6, (Addison-Wesley Publishing Company, Reading, Mass.), 1974. Pp. xviii + 382. Price $ 19.50.

The book under review contains an exposition of modern methods of tackling integral equations. The imbedding methods developed by Richard Bellman and the authors has had a big impact in analytic and computational methods of solution of a variety of problems in physical and biological sciences. It is but appropriate that the authors who have significantly contributed to these areas have come foreward with a monograph which isolates and unifies the techniques of solving integral equations.

The monograph is divided into eight chapters. The first chapter deals with the solution of the Fredholm equation

$$u(t) = f(t) + \int_0^a k(t, y)\, u(y)\, dy$$

where

$$k\ (t, y) = \sum_{m=1}^{M} g_m(t)\ h_m(y).$$

The method is illustrated by a good number of examples backed up by a listing of tested computer programmes. This is followed by a discussion on resolvents for a class of initial value problems. The next chapter (4) deals with kernals having the form

$$k(t, y) = k(|\,t - y\,|) = \int_0^x e^{-|t-y|/z}\ w(z)\, dz.$$

The results that have been obtained in the classical problems of radiation transfer are thrown in a general setting and the authors bring out the possibility of dealing directly with certain functionals of the solution. Chapter 5 deals with the special technique of blending the imbedding method with other suitable techniques like quasi linearization. Chapter 6 contains a good treatment of the resolvents when the kernals are slightly more general. This is followed by a presentation of general techniques of solving non-linear integral equations. Chapter 8 deals with integral equations involving a parameter and the authors provide an exposition of the so-called 'parameter imbedding'. This is followed by a comprehensive treatment of radiative transfer. The final chapter contains a very short account of applicability of the methods to dual integral equations.

The authors have put in considerable effort to isolate the methods from the various physical problems that have been solved during the last decade and present them in a cogent fashion. The book is a valuable contribution to the theory of integral equations.

S. K. SRINIVASAN.

Multiple Scattering Processes Inverse and Direct. By H. H. Kagiwada, R. Kalaba and S. Ueno. Editor R. Kalaba. (Published by Addison-Wesley Publishing Company, Inc., Advanced Book Program, Applied Mathematics and Computation Series), 1975. Pp. xvi + 336. Price $ 21.50.

This volume comes from a group of experts in hte field of transport of radiation and astrophysics who have been engaged for more than a decade in forging new and effective mathematical tooling for the treatment of key problems in transport theory, or in general, for analyzing boundary value problems, with a view to utilize effectively the vast inherent capabilities of the modern digital computer. An initial glimpse into these ideas was provided by the book *Invariant Imbedding and Radiative Transfer in Slabs of Finite Thickness,* by R. E. Bellman, R. E. Kalaba and M. C. Prestrud (American Elsevier, New York, 1963). The present book takes us further to the forefront of a fascinating area of research in this field available till now only in research journals. I congratulate the authors in bringing forth such a lucid and self-contained book, which after laying the foundations of the imbedding approach, develops into a remarkable textbook, wherein theoretical and computational procedures are combined with equal emphasis.

Since multiple scattering is a central feature in many fields such as meteorology, diffusion of radiation, radiative transfer, neutron transport and heat transfer (to mention a few), this textbook is a must for graduate students in these fields. Also, this text provides a gourmet's dish to the applied mathematician interested in boundary value problems, integral equations, system identification problems, etc.

Chapter one introduces basic concepts in one-dimensional transport, and derives the key equations, the integral and differential equations satisfied by

J (t, x) ; $u(t, x)$, the two types of source functions, $r(x)$ the reflection function and the internal intensities $m(t, x)$, $n(t, x)$ at any point t, of the right moving and left moving fluxes for a rod of length x. These are derived both analytically and using the imbedding principle and particle counting techniques. The famous x and Y functions of Chandrasekhar are set forth in a simple context here. Cauchy systems of equations for all these quantities are obtained in elegant ways.

In Chapter Two, the radiative transfer equation relating to multiple scattering processes in an isotropically scattering slab is described in detail, including the function P $(t, x; v)$ which represents the probability that a light quantum absorbed at t finally emerges at an angle v. The equations governing the source function J (t, x, u), X (x, u), Y (x, u) and I $(t, x; v)$ (the internal intensity function) are arrived at by imbedding as well as by other analytical methods. The Cauchy system, for the coupled set of equations for these functions, is solved numerically using simple quadrature for different situations. The results of the numerical computations, including the FORTRAN programs, are given in sufficient detail. These illustrate the advantages of using these imbedding techniques in handling boundary value problems.

Chapter Three is a welcome departure from the usual treatments in radiative transfer theory. It deals with in great detail, the function $b(t, x, v)$ and $h(t, x, v)$ which relate to the intensities for omnidirectional incident flux at the top and bottom. Similarly $\phi(t, x)$ which is related to the source function by equation (3.16) is added to the armory of new functions. The upshot of these new developments is that we obtain the usual quantities, namely, the transmission function T $(x, v; u)$, the reflection function R $(x, v; u)$, and other functions like J $(x, v; u)$, X (x, v); Y $(x; u)$ given by a set of algebraic equations involving b and h. This is a great advantage. The Cauchy system of equations governing b and h are obtained and detailed numerical methods and computer programs illustrate the types of calculation involved.

Chapter Four deals with homogeneous slabs of an atmosphere containing emitting sources of different types. Initial value differential equations for computing the internal and emergent intensities and all other relevant quantities are derived and illustrated with computer program details in a very lucid manner. The second half of Chapter Four, after explaining the quasilinearization methods, goes into details of estimating the parameters of the source distribution for various profiles.

Chapter Five describes methods of treating the inhomogeneous media, while Chapter Six is devoted to isotropically scattering slabs bounded by reflectors of the Lambert law type or by specular reflectors. Ingenious methods such as replacing the reflector at the bottom by an equivalent absorbing slab are presented and the equivalence relations exhibited in great detail with a view to obtain the internal, as well as emergent fluxes, and other relevant information of physical interest including estimation of parameters, by initial value methods assisted by computer codes that considerably lighten the computational task.

A number of well-graded examples, given at the end of each chapter, will provide the student a nice itimacy with the physical basis of several functions such as the b and h functions and in addition to this, lead him to appreciate the vital gains in using the approach of imbedding analysis for the boundary value problems (both direct and inverse) and for computing the kernel functions for integral equations. A good bibliography is provided at the end of each chapter.

This is a welcome addition to the list of books published by Addison-Wesley in this series, and I may suggest that they might consider bringing forth a cheaper Asian edition to make this available to an extensive audience.

R. VASUDEVAN.

Fungi of India (Supplement to the List of Indian Fungi, 1962–72). By K. G. Mukerji and R. C. Juneja. (Emkay Publications, Delhi)., 1974. 27·0 × 20·2 cm. Pp. 1–224. Price Rs. 50·00.

Two disciplines of Fungi which are related are Mycology and Plant Pathology. While Fungi in general are mostly destructive because of the diseases they cause, some of them are useful too. Because of their great economic importance, the list of Fungi of India have been compiled by E. J. Butler and G. R. Bisby, up to 1930 and the supplements by B. B. Mundkur (1931–1938), K. Ramakrishnan and C. V. Subramanian (1938–1951), C. V. Subramanian and K. Ramakrishnan (1952–1956), R. N. Tandon and Sudhirchandra (1957–1962) and the present work by K. G. Mukerji and R. C. Juneja from 1962–1972. In all, 924 genera and 3,435 species, recorded during 1962–1973, are compiled in the present work. After an alphabetical arrangement of the genera, every name is followed by an author. The authorities of the genera are based on the original publications and according to Ainsworth and Bisby (*Dictionary of Fungi*, 6th ed., 1971). After each generic name, the systematic position of the taxon is indicated and the large number of references cited, 1687 in all, are useful for easy consultation. At the end of the book there is a list of generic names of hosts and also the substrata along with the fungi reported on them. It is thus a very comprehensive and useful work, thus bringing the list of Fungi up to 1972. The authors themselves have stated that a consistent application of the *Rules of Nomenclature* has not been possible while preparing the index. An errata is appended for accurate reference.

On the whole it is a very commendable work on the list of Fungi of India and therefore, must find a place as a very valuable and useful reference work in all Agricultural Universities and Departments where studies on Fungi are carried out in a detailed and extensive manner.

K. SUBRAMANYAM.

Information Retrieval Systems: *Classification and Indexing in Science* (3rd Edition, Completely revised). By B. C. Vickery. (Butter-worths, London), 1975. Pp. 228. Price $ £ 5.50 net in U.K. only.

Kowledge ceaselessly advances. With scientific and technical knowledge, above all others, that advance is ceassessly accelerating daily in volume. To make this knowledge usable, large numbers of detailed papers and reports must be indexed by subject. Machines are coming into use to aid selection, but behind every index or machine, must be an analysis of the subject-matter. Classification is the technique of subject analysis and the modern classification uses combinatory methods for subtler and more flexible terms than the traditional tree of knowledge.

The book in its third revised edition describes and discusses the construction of classification schedules and their use in various types of indexing systems—alphabetical and systematic—and to examine the emergence of similar trends in mechanical selection.

The introductory chapter illustrates the fact that classification is used in all forms of tool for information retrieval, to explain its value, to outline the structural features of subject classifications and to indicate some of the problems that arise in the construction.

The classification of a subject field is analysed in the second chapter. Here are discussed subjects such as, facets within each field, grouping terms within each facet; representation of entities by compounding characteristics, etc.

"Classification for Arrangement" is discussed in the third chapter. It analyses the problems of classified arrangement. Typical example of the arrangements of books in library shelves and of abstracts in a bibliographical publication are presented.

Chapter four emphasises the importance of Notation for the Classified Catalogue. It presents a *condensed account* of some basic features of notation.

The aim of modern information retrieval is a fully flexible system which should provide: (1) specific reference to each subject, (2) reference to and from any of its compound terms; and (3) to some degree of reference to each term from terms coordinate in array to it. This fact is well reflected in chapter five of the book.

Chapter six deals with an alternative method of indexing, developed over the past few decades, *the postcoordinate approach.*

To supplement the above, there are four appendices, namely, (A) Historical aspects of the classification of science; (B) Examples of faceted classifications; (C) Categories and (D) The classification of chemical substances.

The first and second edition (enlarged) of this work appeared under the same title in 1958. In the present third edition, the text has been revised and updated. The classification of a subject field presented in the second chapter has been expanded to include new examples. The material on indexing chemicals has been removed, but a new-historical appendix on the classification of chemical substances has been added.

All the bibliographic references appended to each chapter have been considerably revised. However a small flaw has crept in, in this otherwise well presented book. The author has failed to link bibliographic references appended to each chapter, directly with the text, *i.e.,* to convert them where appropriate into citations. The reviewer's experienced doubt *at times* as to which of the items are relevant to a particular point. For instance, in which chapter/section of the "Prolegomena" listed at the end of the chapter on "the need for classification" can we find the definition that "Traditionally subject fields have been equated with main classes and defined as "the fairly....knowledge.", referred on page 11 of this chapter.

Sometimes there is no bibliographic references to support a textual description. For instance, on pages 6 and 7, there appears COSATI subject category list and a reference to this has been made on page 4, but in the bibliography for this chapter, there is no reference to COSATI at all.

While the facts behind this study of classification and indexing are not original, except in so far as they are derived from some of the author's earlier works and articles, the manner of treatment and correlation of the facts can justifiably be regarded as throwing new light on the subject.

It is worth mentioning that the author acknowledges with profound respect the influence of Dr. S. R. Ranganathan, the doyen of library science in India as "the columinous writings of S. R. Ranganathan have influenced me greatly, as will be obvious and I had fruitful talks and correspondence with him".

Special Libraries and Information Workers would find it of immense help when they have to design indexing systems to meet the needs of their users. It is well produced, with attractive typography and layout and is an essential tool in the library. Unfortunately the price will restrict this book to very few purchasers other than libraries.

T. K. S. IYENGAR.
G. S. R. RAO.

Collected Papers of Sir Harold Jeffreys on Geophysics and Other Sciences (Vol. 4). *Dissipation of Energy and Thermal History.* Edited by Sir Harold Jeffreys and Birtha Swirles (Lady Jeffreys) (Gordon and Breach, Science Publishers, 42 William IV Street, London, W.C. 2), Pp. xviii + 523. Price £ 19.10.

This is the fourth of six volumes of the collected papers of Sir Harold Jeffreys edited by Sir Harold and Lady Jeffreys. The papers reproduced in this volume have been grouped into five sections: (1) Dissipation in the Earth and Planets, (2) Radioactivity and Geological Time, (3) The Thermal History of the Earth, (4) The Origin of the Earth's Surface Features and (5) Thermal Instability.

The first section contains, among others, papers dealing with the viscosity of the Earth, Rock Creep and Tidal Friction. The paper on "Tidal Friction in Shallow Seas" is particularly interesting.

The second section consists of four papers which use the process of radioactivity to obtain an estimate for the age of the Earth.

The next group contains eleven papers dealing with the Thermal History of the Earth. Some of these papers are very interesting, and some even entertaining! The first paper is a serious criticism of Prof. Joly's theory of Earth History. (In his book 'The Surface History of the Earth' Prof. Joly had offered a theory of the thermal history of the Earth based on the hypothesis that so much heat is generated in the interior by radioactivity as to lead to widespread fusion!) The 1928 *Phil. Mag.* paper is very interesting because it shows that Sir Harold's disagreement with Prof. Joly is not a 'personal' one but deals with the methodology of theoretical physics. To illustrate this I quote a sentence from Sir Harold's paper "The question is essentially one of theoretical physics: given a set of assumptions to investigate their consequences in the light of physical laws and to compare the results with observations........... The onus of proof in a question of theoretical physics is on the advocate of the theory, not on its opponents....". As Sir Harold says in a footnote (1969) there is as much need to state these scientific principles today as there was in 1928!

The fourth section has nine papers dealing with the origin of the surface features of the Earth. In these the formation of mountains as a consequence of thermal contraction is considered, the quantity of sodium in the ocean is used to suggest an average thickness of the sediments and fault formation is considered.

The last group of seven papers deals with convective thermal instability. The Benard–Rayleigh theory of instability of a fluid heated below is reconsidered with revised boundary conditions. It is suggested that the origin of the continents may be due to such a convective instability in an originally liquid planet.

These papers on very diverse subjects span nearly six decades. Among the "new breed" of physicists it is difficult to find as applied mathematician with such a wide spectrum of interests as Sir Harold. Having said this I must add that this book will be of very limited use to a student of geophysics because it is very poorly annotated in the light of later work. As an addition to any library, however, it will be very useful.

G. SRINIVASAN.

5887/76. Published by S. R. S. Sastry, Manager, Current Science Association, Bangalore-560006
and Printed at The Bangalore Press, Bangalore-560018.

PHOSPHAZENES : A GROUP OF PHOSPHORUS–NITROGEN COMPOUNDS

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THE reaction of ammonium chloride and phosphorus pentachloride is complex[1] and gives rise to oligomeric chlorocyclophosphazenes (phosphonitrilic chlorides), $(NPCl_2)_n$, and also several linear species containing $(NPCl_2)$ units end-stopped

$$n\,NH_4Cl + n\,PCl_5 \longrightarrow (NPCl_2)_n + 4n\,HCl \qquad (1)$$

by the elements of HCl and/or PCl_5. Although the first member of the homologous series, hexachlorocyclotriphosphazatriene, $N_3P_3Cl_6$, was isolated by Liebig[2] in 1834, characterisation of higher homologues ($n = 4$ to 7) was only achieved in the latter part of the nineteenth century[3]. The fundamental studies of Stokes on chlorocyclophosphazenes and their ammonolysis and hydrolysis reactions[4] provided the basis for the modern development of the subject.

In the last two decades, there has been considerable progress in many aspects of phosphazene chemistry. In particular, a great deal of attention has been paid to the chemical reactions of the hexachloride, $N_3P_3Cl_6$, to spectroscopic studies and molecular structure, to the nature of bonding in cyclic phosphazenes and to the technological development of cyclic oligomers and open-chain polyphosphazenes. There are several excellent reviews on these and other aspects of phosphazene chemistry[5-12]. In this article, a brief introduction to some of the significant features of the subject is given as well as an account of developments in Bangalore and other laboratories in the last few years. Actual and potential applications of cyclophosphazenes and phosphazene polymers are noted.

BASIC CHEMISTRY

(*a*) *Aminolysis Reactions*

The reaction of the hexachloride $N_3P_3Cl_6$, with primary, secondary and tertiary amines has received a great deal of attention[1,8,9]. Replacement of the chlorine atoms of $N_3P_3Cl_6$ by amino groups may proceed by two different routes—geminal and nongeminal (Fig. 1). The nongeminal mode of replacement permits the possibility of *cis-trans* isomerism at the bis, tris and tetrakis stages of replacement. This type of isomerism depends on the disposition of groups with respect to the plane of the ring. In most reactions, both geminal and nongeminal replacement patterns are found although usually one of them predominates. Table I summarises the results obtained for a number of amines.

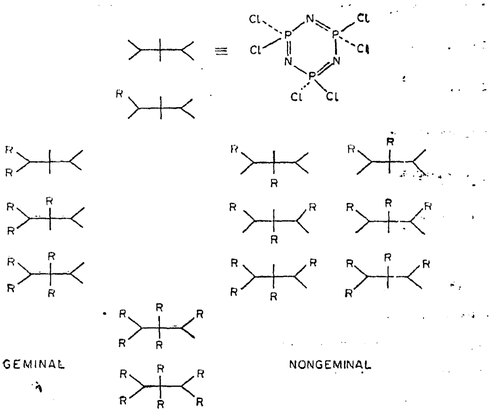


FIG. 1. The geminal and nongeminal pathways in the replacement of chlorine atoms from $N_3P_3Cl_6$ (R groups are attached to P atoms ; chlorine and ring nitrogen atoms are not shown).

Secondary amines react predominantly by the nongeminal mode of replacement although significant quantities of geminal tris-compounds can be isolated in aromatic reaction media[13]. The relative proportion of *cis*- and *trans*- nongeminal isomers varies. The major *bis*- and *tris*-aminocyclophosphazenes formed usually have *trans*-structures whereas the structure of the tetrakisamino derivatives appears to be dominated by steric considerations [$N_3P_3Cl_2(NMe_2)_4$ and $N_3P_3Cl_2(NEt_2)_4$ have *cis*[14]- and *trans*[15]- structures respectively and only minor quantities of their stereo-isomers are obtained]. A "*cis*" effect[16] was proposed to account for the product distribution observed for the reactions of the hexachloride with dimethylamine[14] and piperidine[16]. Recent kinetic data[17] suggest that a substituent solvating effect may also contribute to the preponderance of *trans*- nongeminal *bis*-isomers in secondary amine reactions.

TABLE I

Chlorine atom replacement patterns in the reactions of $N_3P_3Cl_6$ with amines

Amine	Replacement pattern
Ammonia	Geminal
Methylamine	Mainly nongeminal + geminal
Ethylamine	Mainly nongeminal + geminal
Isopropylamine	Nongeminal + geminal
tert-Butylamine	Geminal
Benzylamine	Nongeminal + geminal
Aniline	Mainly geminal + nongeminal
Dimethylamine	Mainly nongeminal + geminal
Diethylamine	Mainly nongeminal + geminal
Pyrrolidine	Mainly nongeminal + geminal
Piperidine	Mainly nongeminal + geminal
Aziridine	Geminal
N-Methylaniline	Nongeminal + geminal

Whereas reactions of $N_3P_3Cl_6$ with secondary amines can be rationalised by the assumption of an S_N2 (P) mechanism, analogous reactions with primary amines are more complex and additional mechanistic features assume importance. Ammonia[18] and *t*-butylamine[19] react with the hexachloride, $N_3P_3Cl_6$, to give only products with geminal structures. A proton abstraction mechanism has been suggested to account for this observation (Fig. 2).

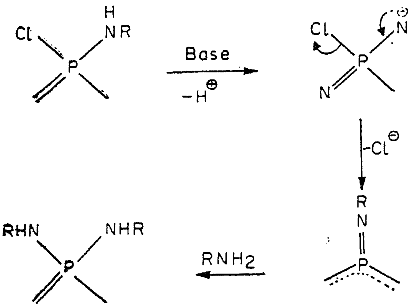


FIG. 2. Proton abstraction mechanism.

Such a mechanism is rendered plausible by the recent isolation[20] of a three-coordinate phosphrous (V) compound, $(Me_3Si)_2NP(=NSiMe_3)_2$. Ethylamine[21] and isopropylamine[22] react initially by the nongeminal mode of replpacement and then by the geminal one (Equation 2). The geminal *tris* and nongeminal tetrakis derivaties have not been isolated

$$N_3P_3Cl_6 \xrightarrow{4NH_2R} \text{nongem-}N_3P_3Cl_4(NHR)_2 \xrightarrow{4NH_2R} \text{gem-}N_3P_3Cl_2(NHR)_4 \qquad (2)$$

although substantial amounts of the former would be expected as intermediates in the formation of geminal tetrakis compounds. In addition to the proton abstraction mechanism, the geminal mode of replacement is probably promoted by the formation of six-membered, cyclic hydrogen-bonded complexes[21] (Fig. 3).

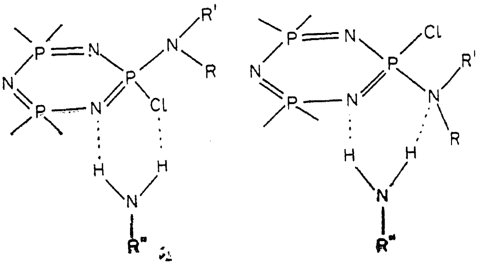


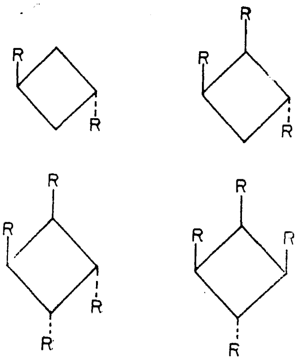
FIG. 3. Possible hydrogen bonded complexes in the reactions of $N_3P_3Cl_6$ with primary amines.

Considerable difficulty is encountered in the isolation[1,9] of pentakisaminochlorocyclotriphosphazatrienes, $N_3P_3Cl(NRR')_5$. The strong electron supply from amino substituents may cause a changeover to a relatively facile heterolysis of the P–Cl bond in an S_N1 type process. Clare and Sowerby[23] have isolated the monochloropentakisdimethylaminocyclotriphosphazatriene, $N_3P_3Cl(NMe_2)_5$, and noted its ready hydrolysis by atmospheric moisture. A stable monochloropentakisamino compound, $N_3P_3Cl(NMe_2)_4[N(CH_2Ph)_2]$, has been reported recently; its stability may be due to steric shielding by the bulky dibenzylamino group[24].

The aminolysis reactions of fluoro- and bromocyclotriphosphazatrienes have not been studied systematically. Some reactions of $N_3P_3F_6$ with ammonia[25] and primray and secondary amines have been reported[26,27]. Fluorodimethylaminocyclotriphosphazatrienes can be prepared by fluorination of the corresponding chloro derivatives[28,29] or by the reaction of the hexakisdimethylamino compound, $N_3P_3(NMe_2)_6$, with antimony trifluoride[23]. The reaction of $N_3P_3Br_6$ with dimethylamine[30] gives the mono, three *bis* and two *tris* derivatives (gem and nongem *trans*). The *cis*-isomer, $N_3P_3Br_3(NMe_2)_3$, has been obtained only from the reaction of $N_3P_3(NMe_2)_6$ and hydrogen bromide in boiling xylene[31]. Formation of a tertrakis derivative was detected (tlc)[30,31] but pure compounds could not be obtained. A preliminary investigation of the reaction of $N_3P_3Br_6$ with ethylamine shows that the general features of the system are similar to those found in the analogous reactions[21] of hexachlorocyclotriphosphazatriene, $N_3P_3Cl_6$ (M. N. Sudheendra Rao, Unpublished results).

Aminolysis reactions of the octachloride, $N_4P_4Cl_8$, and the higher homologues $(NPCl_2)_n$ have received little attention ; the experimental problems associated with the separation of complex mixtures of products and the subsequent difficulties in assigning structures to pure isomers are considerable (there are 33 possible dirivatives of the tetramer). Millington and Sowerby have isolated thirteen products from the reaction[32] of $N_4P_4Cl_8$ with dimethlamine in diethyl ether at — 78° C. The reaction proceeds *via* the non-geminal path ; geminal products were obtained only in poor yields. These workers have also prepared many nongeminal fluorodimethylamino compounds, $N_4P_4F_{8-n}(NMe_2)_n$, by the reaction of $N_4P_4(NMe_2)_8$ with antimony fluoride[33] and by similar reactions using *tris-* and tetrakis-chlorodimethylamino precursors[34].

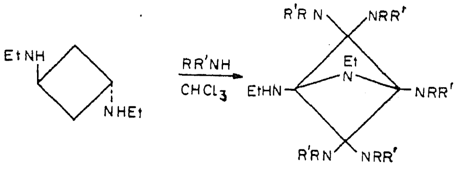
We have recently investigated the reactions of $N_4P_4Cl_8$ with ethylamine[35], *t*-butylamine[36] and N-methylaniline[36]. The reactions of the primary amines have similar features ; mono, bis, tris, tetrakis (ethylamine only) and octakis derivatives can be obtained. Reactions involving higher stoichiometries (particularly 1 : 10 and 1 : 12, $N_4P_4Cl_8$: amine) give only copious quantities of sticky, resinous materials and chloroaminocyclotetraphosphazatetraenes could not be detected[35]. The resins appear to contain tetrameric units and probably arise by cross-linking reactions. The chloroethylamino derivatives isolated have nongeminal structures (Fig. 4). N-Methylaniline reacts with $N_4P_4Cl_8$ to give the derivatives, $N_4P_4Cl_{8-n}(NMePh)_n$ [n = 1, 2 (two isomers, 3, 4 (five isomers), 6] (S. S. Krishnamurthy, R. A. Shaw, M. N. Sudheendra Rao, A. R. Vasudeva Murthy and M. Woods, unpublished results). In contrast to the dimethylamine reaction where only one major *bis*-isomer (2-*trans*-6) is found[32], reaction of $N_4P_4Cl_8$ with N-methylaniline (a much less reactive amine) gives two *bis*-isomers in roughly equal amounts. Crystallographic evidence confirms that one isomer has a 2-*trans*-6 structure and ^{31}P NMR spectroscopy suggests that the other has a 2, 4-structure[36].



R = NHEt

FIG. 4. Structures of $N_4P_4Cl_{8-n}(NHEt)_n$ (n = 2, 3, 4).

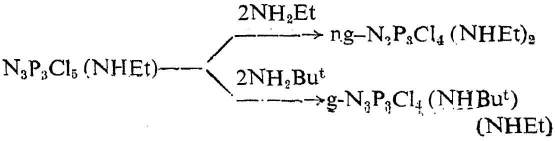
The role of the reaction solvent in cyclophosphazene chemistry is not entirely clear at the moment. Recent work shows that solvents can exert a considerable influence on cyclophosphazene reactions. One of the highlights of a current study has been the isolation of bicyclic phosphazenes from the aminolysis reactions of $N_4P_4Cl_8$ in chloroform. These compounds are probably formed by an intramolecular *trans*-annular nucleophilic replacement reaction (Fig. 5). The crystal structure of $N_4P_4(NMe_2)_5(NHEt)(NEt)$ shows that the original P–N heterocycle retains its phosphazene character but that the P–N bonds at the bridgehead are longer and more phosphazane-like[37]. If these reactions are carried out in diethyl ether[35], the fully substituted cyclophosphazatetraene derivatives, $N_4P_4(NHEt)_2(NRR')_6$ are isolated in good yields (~ 80%).

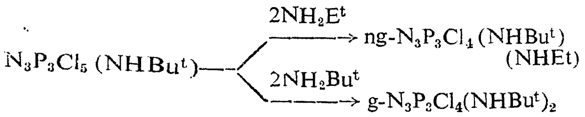


(a) R = R' = Me , (b) R = Et, R' = H

FIG. 5. Formation of bicyclic phosphazenes.

Chlorocyclophosphazenes containing two different amino substituents have also been prepared in order to evaluate the role of substituent and nucleophile in determining the structures of the products[13]. The reactions of $N_3P_3Cl_5(NHEt)$ and $N_3P_3Cl_5(NHBu^t)$ with two equivalents of *t*-butylamine and ethylamine are shown in Scheme 1. The structures





$$N_3P_3Cl_5(NHBu^t) \begin{cases} \xrightarrow{2NH_2Et} ng\text{-}N_3P_3Cl_4(NHBu^t)(NHEt) \\ \xrightarrow{2NH_2Bu^t} g\text{-}N_3P_3Cl_4(NHBu^t)_2 \end{cases}$$

g—geminal ng-nongeminal

of the products obtained from the reactions indicate that the attacking nucleophile dominates the course of these aminolysis reactions[38]. It appears that reactions of the octachloride, $N_4P_4Cl_8$, with primary amines[39], are similarly influenced by the nucleophile. The important role of the nucleophile in many cyclophosphazene reactions has also been confirmed by the recent kinetic studies of Goldschmidt and Licht[17]. Chlorocyclophosphazenes containing both primary and secondary amino substituents have also been prepared and similar conclusions can be drawn[9,24].

It appears that in some reactions the substituent already present in the cyclophosphazene ring can counteract the influence of the incoming nucleophile. At present, the only example of this type is provided by the triphenylphosphazenyl ($NPPh_3$) substituent. Dimethylamine[14] and piperidine[16] react with monosubstituted derivatives, $N_3P_3Cl_5R$, to give nongeminal products, $N_3P_3Cl_4R_2$ ($R = NMe_2$, pip) but with $N_3P_3Cl_5(NPPh_3)$ to give the geminal product, $N_3P_3Cl_4R(NPPh_3)$[40,41].

(*b*) *Alcoholysis and Thioalcoholysis*

The reactions of chlorocyclophosphazenes with alcohols, phenols and thiols have also been studied in some detail[8,9]. Alcohols and phenols react mainly by nongeminal replacement of chlorine atoms. The separation of pure chloroalkoxy (aryloxy) isomers is not easy, particularly when small, straight-chain alcohols are employed. Recently, Schmutz and Allcock[42] have prepared and separated nine trifluoroethoxy derivatives, $N_3P_3Cl_{6-n}(OCH_2CF_3)_n$ ($n = 1$-6) by gas liquid chromatography. Several *n*-butoxy derivatives, $N_3P_3Cl_{6-n}(OBu_n)_n$ ($n = 1, 2, 3, 6$) have also been reported[43].

Thiols react exclusively by a geminal mechanism which has been rationalised in terms of 'hard' and 'soft' acid/base interactions[44,45]. Chlorothioalkoxycyclophosphazenes are obtained by using a large excess of a sodium thiolate and usually only an even number of chlorine atoms is replaced (Equation 3).

$$N_3P_3Cl_6 + NaSR \xrightarrow{\text{organic solvent}} N_3P_3Cl_{6-n}(SR)_n \quad n = 2,4,6 \qquad (3)$$

$$N_3P_3F_6 + NaSEt \longrightarrow N_3P_3F_{6-n}(SEt)_n \quad n = 1\text{-}5 \qquad (4)$$

The hexafluoride reacts with sodium ethanethiolate to give mono, *bis*, *tris*, tetrakis and pentakis ethylthio derivatives but again only compounds with geminal structures are obtained (Equation 4). Derivatives containing -SAr groups in nongeminal dispositions can be obtained by an alternative route[47] (Equation 5).

$$cis - N_3P_3Cl_2(NMe_2)_4 + NaSR \longrightarrow cis - N_3P_3(SR)_2(NMe_2)_4 \qquad (5)$$
$$R = p\text{-}C_6H_4Me,\ Ph$$

Although fully substituted thio-esters, $N_3P_3(SR)_6$, can be prepared[44], the analogous compounds in the tetramer series were not obtained : the only reaction products were geminal tetra-substituted derivatives, $N_4P_4Cl_4(SR)_4$, and organic disulphides, R_2S_2. Cleavage of the tetrameric ring occurred under more forcing reaction conditions[45].

An interesting feature of the chemistry of alkoxycyclophosphazenes $[NP(OR)_2]_{3,4}$ is their ability to rearrange when heated alone[48] (Fig. 6) or in the presence of an alkyl halide[49]. Thio-analogues do not undergo this reaction.

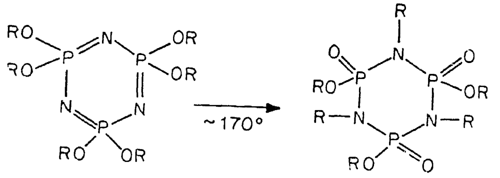


FIG. 6. Alkoxyphosphazene-oxophosphazene rearrangement.

(*c*) *Reactions with Organometallic Reagents*

The hexachloride, $N_3P_3Cl_6$, reacts with phenyl magnesium bromide in diethyl ether to give small quantities of the cyclic hexaphenyl derivative, $N_3P_3Ph_6$ and acyclic phenylated phosphazenyl magnesium complexes from which hydrogen halide derivatives, *e.g.*, $Ph_3P = N - PPh_2 = NH\cdot HX$, ($X = Cl$, Br) can be isolated[50]. Similar reaction with the octachloride, $N_4P_4Cl_8$, gives two cyclic products[51a], $N_4P_4Cl_4Ph_4$, which have the structures shown in Fig. 7. The ring contraction can be explained by a mechanism involving cleavage of the tetrameric ring followed by cyclisation of

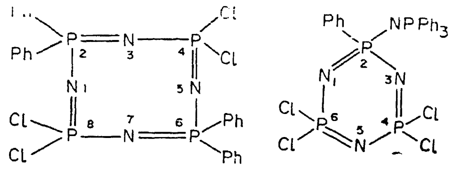


FIG. 7. Products of the reaction of $N_4P_4Cl_8$ with PhMgBr.

the phenylated acyclic species, $Ph_3 P = N - P (Ph Cl) [= N - PPh_2]_2 = N\text{–}Mg\text{–}Br$.

Phenyl lithium reacts with the hexachloride, $N_3P_3Cl_6$ in diethyl ether to give soluble acyclic polymers of low molecular weight[51 b]. The latter possess unreacted phosphorus chlorine bonds and chain branching is evident. The analogous reaction[52] with the hexafluoride, $N_3P_3F_6$, gives the cyclic products, $N_3P_3F_{6-n}\ Ph_n$, $n = 1, 2$ (three isomers) ; nongeminal structures are predominant. The reaction of the octafluoride, $N_4P_4F_8$, with methyl lithium gives methyl, dimethyl, trimethyl, tetramethyl and octamethyl derivatives. The replacement pattern observed is predominantly geminal and has been interpreted in terms of a π-inductive effect[53].

Non-geminal phenylated chlorocyclophosphazenes can be prepared by a cyclisation reaction[54] (Equation 6). Stereoisomers have been obtained for both trimer and tetramer in this reaction.

$$n\, PhPCl_4 + n\, NH_4Cl \longrightarrow (PhNPCl)_n + 4n\, HCl \quad n=3, 4 \qquad (5)$$

Bromophenyl derivatives can be prepared similarly[55].

The Friedel Crafts reaction[56] of the hexachloride, $N_3P_3Cl_6$, with boiling benzene in the presence of aluminium chloride gives the geminal diphenyl derivative, $N_3P_3Cl_4Ph_2$. The tetra- and hexa-phenyl derivatives can be obtained under more forcing conditions. Similar reactions with aminochlorocyclophosphazenes, $N_3P_3Cl_{6-n}\ R_n$ ($R = NMe_2$[57], Pip[58]) indicate that phenylation proceeds most readily at a $\equiv$ PClR group (Fig. 8).

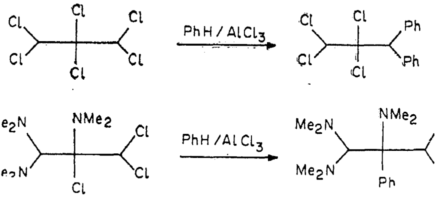


FIG. 8. Friedel Crafts reactions of chlorocyclotriphosphazatrienes.

(d) *Complex and Adduct Formation*

Cyclophosphazenes form many coordination complexes, Lewis acid-base adducts and crystalline inclusion compounds. Allcock[1,8] has reviewed these aspects in detail and two recent papers[59] summarise the subsequent work.

PHYSICAL METHODS OF STRUCTURE DETERMINATION

(a) *Nuclear Magnetic Resonance Spectroscopy*

Nuclear magnetic resonance spectroscopy has emerged as the most powerful tool for elucidating the molecular structures of cyclophosphazene derivatives in solution. The ^1H nmr spectra of some aminochlorocyclophosphazenes illustrate this aspect very clearly. Keat, Ray and Shaw found that *cis-*, *trans-* and geminal-$N_3P_3Cl_3$ $(NMe_2)_3$ isomers can be distinguished by the observation of one, two and three dimethylamino doublets (coupling to phosphorus) respectively in their proton nmr spectra[60]. In addition, the magnitude of the apparent proton-phosphorus coupling constant $^3J^*$ (P–H) helps to distinguish a geminally substituted group ($\equiv PR_2$) from a nongeminal one ($\equiv$ PClR) (R = NHMe, NHEt, NME_2 NMePh). In general, the magnitude of the geminal coupling constant is 3–5 Hz smaller than that of the non geminal one.

In many of the proton spectra of cyclophosphazene derivatives, additional lines or broad humps appear amongst the signals expected from first-order considerations. This phenomenon has usually been termed long range "virtual coupling" and its origin has been discussed elsewhere[61]. Although "virtual coupling" can create problems in the interpretation of many spectra, the extent of "virtual coupling" (or its absence) can sometimes yield structural information (*viz.*, geminal-$N_4P_4Ph_4$ $(NMe_2)_4$ isomers)[62].

The spectra of three chloro-N-methylanilinocyclotetraphosphazatetraenes are shown in Fig. 9 (N-methyl signals only) and illustrate the general points noted above.

The development of sophisticated instrumental facilities and the application of broad band ^1H decoupling have greatly improved the quality and usefulness of the ^{31}P NMR spectra of cyclophosphazenes. The spectra of the three isomers (I, II and III) (Fig. 10) would be of the types AB_2C, $AA'BB'$ and A_2B_2 respectively and are readily identified although if $J/\Delta\nu$ becomes relatively small, it may not be easy to differentiate $AA'BB'$ and A_2B_2 spin systems. The two bis-*t*-butylaminohexachlorocyclotetraphosphazatetraenes, $N_4P_4 (NHBu^t)_2\ Cl_6$, mp 171° and 128°, give rise to symmetrical A_2B_2 and $AA'BB'$ ^{31}P spectra indicating that these isomers have a 2, 6- and 2, 4-disposition of *t*-butylamino groups[39]. The ^{31}P spectra of many amino-derivatives of $N_3P_3Cl_6$ and some selected tetrameric derivatives have been analysed recently and some trends in ^{31}P chemical shifts and P–N–P coupling constants have been discerned[63].

(b) *Infrared Spectroscopy*

Infrared studies of cyclophosphazene derivatives have not proved to be as useful as NMR spectroscopy in determining the disposition of substituent groups, although in some cases it is possible to distinguish geminal and non-geminal isomers in the trimeric system[64]. IR spectroscopy can provide some information on the nature of skeletal bonding. The spectra of cyclophosphazene derivatives exhibit a broad absorption band in the range 1150–1450 cm^{-1} which has been attributed[65] to a degenerate ring stretching vibration, ν (P = N). Typical values are shown in Table II and illustrate the importance of ring size, electronegativity, steric effects, etc.

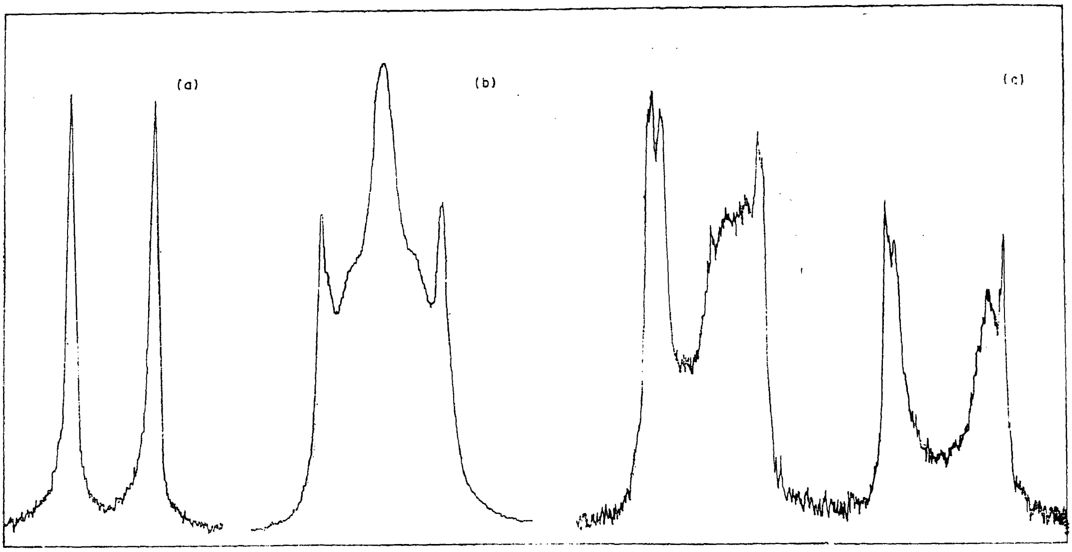
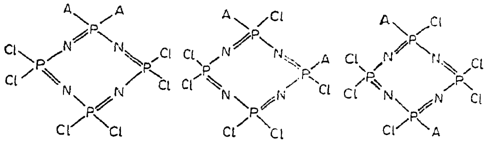


FIG. 9. ^{1}H NMR spectra (CDCl$_3$, 100 MHz) of (*a*) 2, 2, 6, 6,-$N_4P_4Cl_4$ (NMePh)$_4$. (*b*) 2, 4, 6, 8,-$N_4P_4Cl_4$ (NMePh)$_4$. (*c*) 2, 2, 4, 6, 8-$N_4P_4Cl_5$ (NMePh)$_3$ (Frequency separations on the same scale) ; (NMe signals only).



I II III

FIG. 10. Isomeric configurations of $N_4P_4Cl_6A_2$.

(c) Basicity Measurements

The technique of basicity measurements[9,13] developed by Feakins, Shaw and coworkers can be used to distinguish geminal and nongeminal tris- and tetrakis-amino isomers. There is now considerable evidence (including X-ray structures of two protonated compounds)[66] for protonation at a ring nitrogen atom in most cyclophosphazenes. Basicity measurements of cyclophosphazenes containing a triphenylphosphazenyl substituent[41] indicate that protonation can take place *either* at a ring nitrogen atom (Type I) or at the nitrogen atom of the phosphazenyl substituent (Type II), probably because of a difference in conformation of the triphenylphosphazenyl group (prior/or subsequent to protonation). It is believed[13] that in Type I compounds, the N–P bond of the triphenylphosphazenyl group is more or less parallel to the local NPN ring segment whereas in Type II compounds, it is approximately perpendicular. Two recent crystallographic studies[67,68] provide examples of Type I and Type II conformations.

TABLE II

Ring P = N vibrational frequencies in cyclophosphazene (cm^{-1})^a

R	$N_3P_3R_6$	$N_4P_4R_8$
F	1297	1435
Cl	1218	1315
Br	1175	1275
Me	1180	1220^c
Ph	1190	1213
NH_2	1170	1240
NHEt	1180^b	1250^b
NHBu^n	1195	1260
NHC_2H_{11}^{n}	1190	1265
NMe_2	1195	1265

a Reference 6, p. 197.
b Reference 39, *c* Reference 110.

(d) X-ray Crystallography

The structures of a large number of cyclophosphazene derivatives have been determined by X-ray diffraction. Corbridge has reviwed this work recently[68]. The equality of ring P–N bond lengths in homogeneously substituted cyclophosphazene derivatives and the shortness of these bonds compared with the accepted value (1·77 Å) for P–N single bond have provided good evidence for some kind of π–interaction involving nitrogen and phosphorous atoms. The X-ray crystallographic data have also provided strong evidence for the presence of π–bonding between phosphorus and exocyclic groups such as $-NMe_2$ or –OR. It has been pointed out that the ring P–N bond lengths in a derivative $N_3P_3X_6$ decrease with increasing electronegativity of X. This is also accompanied by a decrease in the angle subtended by the exocyclic substituents at phosphorus, an increase in the ring NPN angle and a decrease in the ring PNP angle. In a heterogeneously substituted compound, the ring P–N bonds are of different lengths. Thus in the geminal diphenyl compound, $N_3P_3Cl_4Ph_2$, there are three different P–N bond lengths[70] with mean values 1·555, 1·578 and 1·615 Å. The analogous fluoro compound[71] and the geminal tetraphenyl[72] compound, $N_3P_3Cl_2Ph_4$, also show three types of ring P–N bonds. These differences can be attributed to greater *d*-orbital contractions at the phosphorus atom bearing the most electronegative substituents leading to a strrengthening of the skeletal π–bonding. The X-ray crystal structures of cyclotetraphosphazatetraenes show that the eight membered P–N ring can adopt one of several different conformations (Table III).

(e) Other Techniques

Nuclear Quardrupole Resonance spectra (^{35}Cl) of some chlorocyclophosphazene derivatives have been reported. A linear relationship has been observed between NQR frequency and P–Cl bond length[73]. Positional isomers may be distinguished by this technique but unambiguous structural assignments to geometrical isomers are not generally possible[74].

It should be possible, in principle, to distinguish geometrical isomers from dipole moment measurements as demonstrated for the *cis* and *trans* isomers of $N_3P_3Ph_3Br_3$[55], $N_3P_3Cl_4(NMe_2)_2$ and $N_3P_3Cl_3(NMe_2)_3$[75]. Such measurements are unsuitable for the identification of chloro (trifluroethoxy) cyclotriphosphazene isomers. This difficulty is presumably due to the neglect of the atom polarizability term and the possible deviations of the ring from planarity[42].

Bonding in Cyclophosphazenes

Bonding in cyclophosphazenes (characterised by a formally unsaturated system) has intrigued a number of investigators. Craig and Paddock have proposed a $d\pi - p\pi$ model in which extensive delocalization of π electrons occurs over the entire ring[76]. The same authors also postulated[77] a π-bonding interaction in the ring plane involving the lone pair of electrons on the skeletal nitrogen atom (π'-bonding). Dewar *et al.* have put forward an alternative model in which π-electron interaction is confined to three-centre P–N–P islands[78]. Detailed quantum mechanical calculations[79] and Faraday effect studies[80] on cyclophosphazenes tend to support the latter model. Doggett[81] has suggested that the two models do not differ fundamentally but only in the choice of parameters. Labarre, Perkins and their respective coworkers have pointed out that *trans*-annular phosphorus—phosphorus bonding may contribute significantly to the stability of the six-membered P–N ring system[82].

PHOSPHAZENE HIGH-POLYMERS

(a) Cyclolinear and Cyclomatrix Polymers

There are three basic approaches to the synthesis of phosphazene based polymers leading to the formation of cyclolinear, cross-linked cyclomatrix and linear type polyphosphazenes[1,8,11,12,83]. The first two types utilize the high thermal stability of cyclotri- and cyclotetraphosphazene rings by linking them through bifunctional organic groups. The formation of cyclolinear polymers is illustrated in Fig. 11.

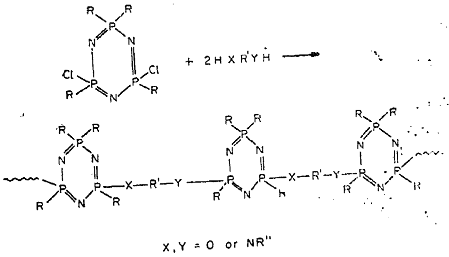


FIG. 11. The formation of cyclolinear polymers.

Cyclophosphazene substrates with more than two active groups (usually halogen) may also react with diols or diamines to give cyclomatrix polymers (Fig. 12). The extensive cross-linking usually found in such polymers is responsible for their insolubility and high melting points.

(b) Linear Polymers

Chlorocyclophosphazenes, $(NPCl_2)_{3-7}$, on heating in the bulk state under non-rigorous experimental

TABLE III

X-ray structural data for cyclotetraphosphazatetraenes

Compound	Ring shape	Mean ring P–N bond length (Å)	Mean ring P–N–P angle (°)	Mean ring N–P–N angle (°)	Ref.
$N_4P_4F_8$	Planar	1·51	120·3	119·4	91
$N_4P_4Cl_8$ (K)	Intermediate Boat-saddle	1·57	131·3	121·2	92
$N_4P_4Cl_8$ (T)	Chair	1·56	135·6	120·5	93
$N_4P_4Br_8$	Intermediate Boat-saddle	1·575	131·0	120·1	94
$N_4P_4F_6Me_2$ (2, 2, 4, 4, 6, 6 : 8, 8)	Saddle	1·525	145·0	121·5	95
$N_4P_4F_4Me_4$ (2, 2, 4, 4: 6, 6, 8, 8)	Saddle	1·56	134·6	121·7	96
$N_4P_4F_4(NMe_2)_4$ 2-*cis*-4-*trans*-6-*trans*-8	Chair	1·55	139·5	121·3	97
$N_4P_4F_4(NMe_2)_4$ 2-*trans*-4-*cis*-6-*trans*-8	Saddle	1·557	134·7	122·0	98
$N_4P_4Cl_4Ph_4$ (2, 2, 4, 4: 6, 6, 8, 8)	Saddle	1·553, 1·591	128·8– 135·4	120·0	99
$N_4P_4Cl_4Ph_4$ 2-*cis*-4-*cis*-6-*cis*-8	Irregular crown	1·57	137·5	121·0	100
$N_4P_4Cl_4Ph_4$ 2-*cis*-4-*trans*-6-*trans*-8	Chair	1·57	135·4	120·0	101
$N_4P_4Cl_6$ (NMe)$_2$ 2-*trans*-6	Chair	1·569	134·2	120·6	102
$N_4P_4Cl_4(NMe_2)_4$ 2-*cis*-4-*trans*-6-*trans*-8	Hybrid crown-saddle	1·556	136·8	121·1	103
$N_4P_4Cl_2(NMe_2)_6$ 2-*trans*-6	Chair	1·57	135·4	120·7	104
$N_4P_4Me_8$	Hybrid boat-saddle	1·60	131·9	119·8	105
$N_4P_4Ph_8$	Hybrid boat-saddle	1·59	127·8	119·8	106
$N_4P_4(OMe)_8$	Saddle	1·57	132·0	121·0	107
$N_4P_4(NMe_2)_8$	Hybrid boat-saddle	1·58	133·0	120·0	108
$N_4P_4(NCS)_8$	Chair	1·543	140·0	120·2	109

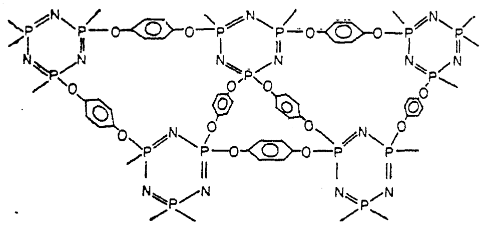


FIG. 12. A phosphazene cyclomatrix polymer.

conditions give an elastomeric polymer called "inorganic rubber" which is highly cross-linked and insoluble in organic solvents. This polymer is hydrolytically unstable and becomes brittle on prolonged exposure to moist air. Efforts to overcome this problem by replacement of chlorine atoms with hydrolytically stable organic groups have been unsuccessful because the reactions are very slow and complete replacement of chlorine atoms inside the cross-linked matrix is virtually impossible. Allcock and coworkers[84] have shown that "poly (dichlorophosphazene)" free from cross-links can be obtained by carrying out the polymerization of $N_3P_3Cl_6$ under carefully controlled conditions and by limiting the conversion to $\sim$ 70%. They have recently established that traces of water, function as a powerful catalyst whereas PCl_5 is a powerful inhibitor for the polymerization reaction[85]. The mechanism of the polymerization reaction is not yet fully understood.

Poly (dichlorophosphazene), free from cross-linking[1,85], is soluble in organic solvents and reacts readily and completely with a wide variety of nucleophilic reagents (Fig. 13). Copolymers and terpolymers containing different substituents can be prepared[11]. Generally the homopolymers are flexible film-forming thermoplastics whereas the copolymers are elastomeric in nature. Polymers with different physical characteristics can be obtained by varying the substituents. Table IV summarises the properties of some selected linear polyphosphazenes. One of the most attractive features of the phosphazene linear polymeric system is the prospect of designing polymers for specific applications by the judicious choice of suitable substituent groups.

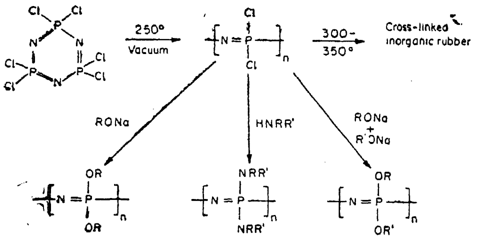


FIG. 13. The reactions of poly (dichlorophosphazene).

Potential Applications

Hexa-ammonocyclotriphosphazatriene, $N_3P_3(NH_2)_6$, is found to be a good fertilizer for certain crops as it can supply both nitrogen and phosphorus in a concentrated form[86]. The aziridinocyclophosphazenes have been used as chemosterilant insecticides (under the trade name "apholate")[87]. The alkoxycyclophosphazenes, in particular the *n*-propoxy derivatives, are excellent flame retardants for viscose rayon[88]. Phosphazene cyclomatrix polymers are amenable to conventional "curing" processes. In has been suggested that they may find use in high temperature aerospace applications and as heat-resistant coatings for electrical components[89]. Linear polyphosphazenes containing fluoroalkoxy groups are resistant to acids, alkales, and hydrocarbon fuels and retain their flexibility over a wide temperature range. They are being developed for fabricating lip seals, gaskets and fuel hoses serviceable at low temperatures (− 40° C)[11]. Poly (alkoxyphosphazene) derivatives have good flame retarding properties and may find extensive use as textile impregnating agents and fire resistant foams. Polyphosphazenes also show promise as body implantation plastics[90].

TABLE IV

Properties of some linear polyphosphazenes[1,11]

Compound	Molecular weight	Intrinsic viscosity dl/g	T_g (° C)[c]	Solubility[d]
$(NPCl_1)_n$	1–2 × 10^6 [a,e]	2·40	−63	Benzene
$[NP(OMe)_2]_n$	6,40,000[a]	1·22	−76	Methanol
$[NP(OCH_2CF_3)]_n$	1,700,000[a]	2·70	−66	Ketones
$[NP(OCH_2CF_3)(OCH_2C_3F_7)]_n$	>6 × 10^6 [a]	2·40	−77	Freons
$[NP(OPh)_2]_n$	3,700,000[a]	2·70	5·5	THF
$[NP(OC_6H_4Cl\text{-}m)]_n$	3·89 × 10^6 [a]	..	−24	Chloroform
$[NP(NHPh)_2]_n$	1,650,000[a]	1·44	91	Benzene
$[NP(NEt_2)(NHMe)]_n$	230,000[b]	1·41	8	THF

a Light scattering.
b Ultracentrifugation.
c By differential thermal analysis or differential scanning calorimetry.
d At room temperature.
e Not very accurate owing to slow cross-linking in solution.

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1. Allcock, H. R., *Phosphorus-Nitrogen Compounds,* Academic Press, New York, 1972.
2. Liebig, J., *Ann. Chem.,* 1834, 11, 139.
3. Stokes, H. N., *Amer. Chem. J.,* 1897, **19**, 782.
4. Stokes, H. N., *Ibid.,* 1895, 17, 275; *Ibid.,* 1896, **18**, 629.
5. Shaw, R. A., Fitzsimmons, B. W. and Smith, B. C., *Chem. Rev.,* 1962, **62**, 247.
6. Paddock, N. L., *Quart. Rev., Chem. Soc.,* 1964, 18, 168.
7. Sowerby, D. B., "Phosphonitriles", In: *MTP International Review of Science,* Inorganic Chemistry Series, I, 1972, 2, 117.
8. Allcock, H. R., *Chem. Rev.,* 1972, 72, 315.
9. Keat, R. and Shaw, R. A., In: *Organic Phosphorus Compounds,* Kosolapoff, G. M. and Maier, L. (Ed.), Interscience, New York, 1973, Vol. 6, 833.
10. Goldschmidt, J. M. E., "The Cyclophosphazenes", In: *Analytical Chemistry of Phosphorus Compounds,* Halmann, M., (Ed.), Wiley—Interscience, New York, 1972, 13, 523.
11. Singler, R. E., Schneider, N. S. and Hagnauer, G. L., *Polymer Eng. and Sci.,* 1975, **15**, 321.
12. Kireev, V. V., Kolesnikov, G. S. and Raigorodskii, I. M., *Russ. Chem. Rev.* (Eng. Transl.), 1969, **38**, 667.
13. Shaw, R. A., *Z. Naturforsch.* (In press).
14. Keat, R. and Shaw, R. A., *J. Chem. Soc.,* 1965, p. 2215.
15. Lehr, W. and Rosswag, N., *Z. anorg. Chem.,* 1974, **406**, 221.
16. Keat, R. and Shaw, R. A., *J. Chem. Soc.* (*A*), 1966, p. 908.
17. Goldschmidt, J. M. E. and Licht, E., *J. Chem. Soc. Dalton,* 1972, p. 728; *Ibid.,* 1972, p. 732.
18. Feistel, G. R. and Moeller, T., *J. Inorg. Nucl. Chem.,* 1967, 29, 2731; Lehr, W., *Z. anorg. Chem.,* 1967, 356, 18.
19. Das, S. K., Keat, R., Shaw, R. A. and Smith, B. C., *J. Chem. Soc.,* 1965, p. 5032.
20. Scherer, O. J. and Kuhn, N., *Chem. Ber.,* 1974, **107**, 2123; Niecke, E. and Flick, W., *Angew. Chem. Internat. Edn.,* 1974, 13, 134; Pohl, S., Niecke, E. and Krebs, B., *Angew. Chem. Internat. Edn.,* 1975, 14, 261.
21. Das, R., Shaw, R. A., Smith, B. C. and Woods, M., *J. Chem. Soc. Dalton,* 1973, p. 709.
22. Das, S. K., Keat, R., Shaw, R. A. and Smith, B. C., *J. Chem. Soc.* (*A*), 1966, p. 1677.
23. Clare, P. and Sowerby, D. B., *J. Inorg. Nucl. Chem.,* 1974, **36**, 729.
24. Hasan, M., Shaw, R. A. and Woods, M., *J. Chem. Soc. Dalton,* 1975, p. 2202.
25. Roesky, H. W. and Niecke, E., *Inorg. Nucl. Chem. Letters,* 1968, **4**, 463.
26. Glemser, O., Niecke, E. and Roesky, H. W., *J. Chem. Soc. Chem. Comm.,* 1969, p. 282.
27. Chivers, T., Oakley, R. T. and Paddock, N. L., *J. Chem. Soc.* (*A*), 1970, p. 2324.
28. Green, B. and Sowerby, D. B., *J. Chem. Soc.* (*A*), 1970, p. 987.
29. —, — and Clare, P., *Ibid.,* 1971, p. 3487.
30. Stahlberg, R. and Steger, E., *J. Inorg. Nucl. Chem.,* 1966, **28**, 684; *Ibid.,* 1967, **29**, 961.
31. Nabi, S. N., Shaw, R. A. and Stratton, C., *J. Chem. Soc. Dalton,* 1975, p. 588.
32. Millington, D. and Sowerby, D. B., *Ibid.,* 1972, p. 2035.
33. — and —, *Ibid.,* 1974, p. 1070.
34. — and —, *Ibid.,* 1973, p. 2649.
35. Keat, R., Krishnamurthy, S. S., Sau, A. C., Shaw, R. A., Vasudeva Murthy, A. R. and Woods, M., *Ibid.* (In press).
36. —, —, —, —, Sudheendra Rao, M. N., Vasudeva Murthy, A. R. and Woods, M., *Z. Naturforsch.,* 1974, **29 B**, 701.
37. Cameron, T. S., Mannan, Kh., Krishnamurthy, S. S., Sau, A. C., Shaw, R. A., Vasudeva Murthy, A. R. and Woods, M., *J. Chem. Soc. Chem. Comm.,* 1975, p. 975.
38. Keat, R. and Shaw, R. A., *Angew. Chem. Internat. Edn.,* 1968, **7**, 212.
39. Sau, A. C., *Ph.D. Thesis,* I.I.Sc., Bangalore, 1976.
40. Biddlestone, M. and Shaw, R. A., *J. Chem. Soc. Dalton,* 1973, p. 2740.
41. —, Nabi, S. N. and Shaw, R. A., *Ibid.,* 1975, p. 2634.
42. Schmutz, J. L. and Allcock, H. R., *Inorg. Chem.,* 1975, **10**, 2433.
43. Zeleneva, T. P., Antonov, I. V. and Stepanov, H. I., *J. Gen. Chem.* (U.S.S.R.), 1973, **43**, 1000.
44. Carroll, A. P. and Shaw, R. A., *J. Chem. Soc.* (*A*), 1966, p. 914.
45. —, —, and Woods, M., *J. Chem. Soc. Dalton,* 1973, p. 2736.
46. Niecke, E., Glemser, O. and Roesky, H. W., *Z. Naturforsch.,* 1969, **24 B**, 1187.
47. Woods, M., *Ph.D. Thesis,* London University, 1969.
48. Fitzsimmons, B. W., Hewlett, C. and Shaw, R. A., *J. Chem. Soc.,* 1964, p. 4459.
49. —, — and —, *Ibid.,* 1965, p. 7432.
50. Biddlestone, M. and Shaw, R. A., *J. Chem. Soc.* (*A*), 1969, p. 178; *J. Chem. Soc. Dalton,* 1975, p. 2527.
51. (*a*) — and —, *Ibid.,* 1970, p. 1750; (*b*) — and —, *Phosphorus,* 1973, 3, 95.
52. Allen, C. W. and Moeller, T., *Inorg. Chem.,* 1968, 7, 2177.
53. Ranganathan, T. N., Todd, S. M. and Paddock, N. L., *Ibid.,* 1973, 12, 317.
54. Grushkin, B., Sanchez, M. G. and Rice, R. G., *Ibid.,* 1964, 3, 623; Humiec, F. S. and Bezman, I. I., *J. Amer. Chem. Soc.,* 1961, 83, 2210; Shaw, R. A. and Stratton, C., *J. Chem. Soc.,* 1962, p. 5004.

55. Moeller, T. and Nannelli, P., *Inorg. Chem.*, 1963, 2, 659; *Ibid.*, 1963, 2, 896.
56. Acock, K. G., Shaw, R. A. and Wells, F. B. G., *J. Chem. Soc.*, 1964, p. 121; McBee, E. T., Okishara, K. and Morton, C. J., *Inorg. Chem.*, 1965, 4, 1672.
57. Das, S. K., Shaw, R. A. and Smith, B. C., *J. Chem. Soc. Dalton*, 1973, p. 1883.
58. —, — and —, *Ibid.*, 1974, p. 1610.
59 Searle, H. T., Dyson, J., Ranganathan, T. N. and Paddock, N. L., *Ibid.*, 1975, p. 203; Calhoun, H. P., Paddock, N. L. and Wingfield, J. N., *Can. J. Chem.*, 1975, 53, 1765.
60. Keat, R., Ray, S. K. and Shaw, R. A., *J. Chem. Soc.*, 1965, p. 7193.
61. Finer, E. G., Harris, R. K., Bond. M. R., Keat, R. and Shaw, R. A., *J. Molecular Spectroscopy*, 1970, 33, 72; Finer, E. G., *Ibid.*, 1967, 23, 104.
62. Biddlestone, M., Bullen, G. J., Dann, P. E., Krishnamurthy, S. S., Shaw, R. A. and Woods, M., *Phosphorus*, 1973, 3, 179.
63. Keat, R., Shaw, R. A. and Woods, M., *J. Chem. Soc. Dalton* (In press).
64. Chapman, A. C., Paine, D. H., Searle, H. T., Smith, D. R. and White, R. F. M., *J. Chem. Soc.*, 1961, p. 1768; Stahlberg, R. and Steger, E., *Spectrochim. Acta*, 1967, 23 A, 2005.
65. —, and Paddock, N. L., *J. Chem. Soc.*, 1962, p. 635.
66. Mani, N. V. and Wagner, A. J., *Acta Cryst.*, 1971, 27 B, 51; Allcock, H. R., Bissell, E. and Shawl, E. T., *Inorg. Chem.*, 1973, 12, 2963.
67. Biddlestone, M., Bullen, G. J., Dann, P. E. and Shaw, R. A., *J. Chem. Soc. Chem. Comm.*, 1974, p. 56.
68. Babu, S., Cameron, T. S., Krishnamurthy, S. S., Manohar, H. and Shaw, R. A., *Z. Naturforsch.* (In press).
69. Corbridge, D. E. C., *The Structural Chemistry of Phosphorus*, Elsevier, Amsterdam, 1974, p. 333.
70. Mani, N. V., Ahmed, F. R. and Barnes, W. H., *Acta Cryst.*, 1965, 19, 693.
71. Allen, C. W., Paul, I. C. and Moeller, T., *J. Amer. Chem. Soc.*, 1967, 89, 6361.
72. Mani, N. V., Ahmed, F. R. and Barnes, W. H., *Acta Cryst.*, 1966, 21, 375.
73. Keat, R., Porte, A. L., Tong, D. A. and Shaw, R. A., *J. Chem. Soc. Dalton*, 1972, p. 1648.
74. Dalgleish, W. H., Keat, R., Porte, A. L., Tong, D. A., Hasan, M. and Shaw, R. A., *Ibid.*, 1975, p. 309.
75. Koopman, H., Spruit, F. J., Van Deursen, F. and Backkers, J., *Rec. Trav. Chim.*, 1965, 84, 341.
76. Craig, D. P. and Paddock, N. L., *Nature*, 1958, 181, 1052.
77. — and —, *J. Chem. Soc.*, 1962, p. 4118.
78. Dewar, M. J. S., Lucken, E. A. C. and Whitehead, M. A., *Ibid.*, 1960, p. 2423.
79. Faucher, J.-P., Devanneaux, J., Leibovici, C. and Labarre, J. F., *J. Molecular Struct.*, 1971, 10, 439; Faucher, J.-P., Labarre, J. F. and Shaw, R. A., *Ibid.*, 1975, 25, 109.
80. Faucher, J. P., Glemser, O., Labarre, J.-F. and Shaw. R. A., *CR Acad. Sci. Paris*, 1974, 279 C, 441.
81. Doggett, G., *J. Chem. Soc., Faradey* II, 1972, p. 2075.
82. Armstrong, D. R., Longmuir, G. H. and Perkins, P. G., *J. Chem. Soc. Chem. Comm.*, 1972, p. 464; Faucher, J.-P. and Labarre, J.-F., *Phosphorus*, 1974, 3, 265.
83. Allcock, H. R., *Chem. in Britain*, 1974, p. 118.
84. — and Kugel, R. L., *J. Amer. Chem. Soc.*, 1965, 87, 4216; Allcock, H. R., Kugel, R. L. and Valan, K. J., *Inorg. Chem.*, 1966, 5, 1709.
85. —, Gardner, J. E. and Smeltz, K. M., *Macromolecules*, 1975, 8, 36.
86. Wakefield, Z. T., Allen, S. E., McCullough, J. F., Sheridan, R. C. and Kohle, J. J., *J. Agr. Food Chem.*, 1971, 19, 99.
87. *Chem. Week*, 1963, Dec. 14, 83.
88. Godfrey, L. E. A. and Schappel, J. W., *Ind. Eng. Chem. Prod. Res. Develop.*, 1970, 9, 426.
89. *Reference*, 1, p. 377.
90. *Chem. and Eng. News*, 1975, June 16, 22.
91. McGeachin, H. M. and Tromans, F. R., *J. Chem. Soc.*, 1961, p. 4777.
92. Hazekamp, R., Migchelsen. T. and Vos, A., *Acta Cryst.*, 1962, 15, 539.
93. Wagner, A. J. and Vos, A., *Ibid.*, 1968, 24 B, 707.
94. Zoer, H. and Wagner, A. J., *Ibid.*, 1972, 28 B, 252.
95. Marsch, W. C. and Trotter, J., *J. Chem. Soc. (A)*, 1971, p. 573.
96. Marsh, W. C. and Trotter, J., *Ibid.*, 1971, p. 569.
97. Millington, D., King, T. J. and Sowerby, D. B., *J. Chem. Soc. Dalton*, 1973, p. 396.
98. Begley, M. J., Millington, D., King, T. J. and Sowerby, D. B., *Ibid.*, 1974, p. 1162.
99. Bullen, G. J. and Dann, P. E., *Acta Cryst.*, 1974, 30 B, 2861.
100. — and Tucker, P. E., *J. Chem. Soc. Dalton*, 1972, p. 1651.
101. Burr, A. H., Carlise, C. H. and Bullen, G. J., *Ibid.*, 1974, p. 1659.
102. Bullen, G. J. and Dann, P. E., *Ibid.*, 1973, p. 1453.
103. — and Tucker, P. A., *Ibid.*, 1972, p. 2437.
104. — and Dann, P. E., *Ibid.*, 1974, p. 705.
105. Dougill, M. W., *J. Chem. Soc.*, 1961, p. 5471.
106. Begley, M. J., Sowerby, D. B. and Tillet, R. J., *J. Chem. Soc. Dalton*, 1974, p. 2527.
107. Ansell, G. B. and Bullen, G. J., *J. Chem. Soc. (A)*, 1971, p. 2498.
108. Bullen, G. J., *J. Chem. Soc.*, 1962, p. 3193.
109. Faught, J. B., *Can. J. Chem.*, 1972, 50, 1315.
110. Searle, H. T., *Proc. Chem. Soc.*, 1959, p. 7.

ION-EXCHANGE PROPERTIES OF HYDROUS CERIC OXIDE II-SORPTION OF SOME METAL ETHYLENE DIAMMINES

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INTRODUCTION

A SURVEY of the literature indicates that ion-exchange properties of hydrous CeO_2 have not been studied[1,2]. Apart from practical utility of the selectivity for the purpose of chemical separation, an understanding of the factors contributing to the selectivity pattern is of considerable theoretical interest. The present study describes an investigation of the sorption behaviour of hydrous ceric oxide using ethylene-diammines of Zn^{2+}, Ni^{2+} and Cd^{2+}. The study includes (*a*) the dependence of the value of q_A and K_d on the pH and the concentration of the exchanging ions and (*b*) selectivity quotients ($K_A{}^B$) at various concentrations of the competing ionic species A and B.

MATERIALS AND METHOD

All the reagents used were of A.R. quality. Hydrous CeO_2 was prepared by adding excess of aqueous ammonia to a saturated solution of ceric ammonium sulphate[3]. The resulting precipitate was washed until the pH of the wash liquid was $\simeq$ 6 and dried at 100–110° C. Ethylenediammine complexes were prepared by dissolving appropriate quantities of the metal salts in water followed by the addition of excess of ethylene diammine and the pH values were adjusted by means of ethylenediammine solution/dil. HCl.

Determination of q_A and K_d

100 mg samples of the hydrous CeO_2 were equilibrated with 20 ml solution of different exchanging complex ions of specified composition (or pH values) and the clean centrifuged supernatants (5 ml) were analysed complexometrically[4]. Knowing the initial and the ekuilibrium concentrations of the metal ions in the external solution, the amount (q_A) sorbed per 100 mg of the exchanger material was calculated. K_d values were calculated by the relation[5]:

$$K_d = \frac{\text{Amount of A in } CeO_2}{\text{Amount of A in solution}} \times \frac{\text{ml solution}}{\text{gm dry resin}}$$

where K_d is the distribution coefficient.

Determination of the Selectivity

Selectivity quotients, $K_A{}^8$, were determined using the relation[6]

$$K_A{}^B = \frac{K_d \text{ for B}}{K_d \text{ for A}}$$

Experimental methods are described below:

For the determination of the selectivity quotient of the competing en-complex ion pairs of Zn^{2+} — Cd^{2+} and Ni^{2+} — Cd^{2+}, a radiometric method[7] was followed. 20 ml solutions of the competing pair, containing equimolar proportions of the metal ions tagged with a drop of radioisotope ^{115}Cd, were equilibrated with 100 mg of the hydrous ceric oxide. From the initial and equilibrium counts the amount of Cd^{2+} ion sorbed was calculated and the amounts of Zn^{2+} and Ni^{2+} sorbed were determined complexometrically[4].

RESULTS AND DISCUSSION

The results are presented in Tables I–IV.

It is well known that sorption of metal ions on hydrous oxide takes place by the mechanism of ion-exchange

$$\geqslant M - OH \xrightleftharpoons{\text{basic medium}} \geqslant M - O^- + H^+ \xrightleftharpoons{X^+} \geqslant M - OX + H^+$$

where M stands for the Ce (IV) ion and X, exchanging metal ions such as Zn^{2+}, Cd^{2+}, etc. Keeping the above mechanism in view, the dependence of sorption capacity on the various experimental factors may be examined.

Effect of Addition of Ethylenediammine (Table I)

Successive additions of ethylenediammine, for the adjustment of pH influences the overall exchange process in the following manner: The increased basicity of the solution facilitates the release of hydrogen ions by exchange and these H^+ ions combine with en molecules producing en-H^+ and en-$H_2{}^{++}$ ions; these cations then compete with the metal complex ions and consequently sorption of the metal ions adversely affected, causing a decrease in q_A and K_d values at higher pH. Thus, the exchange process in the case of $Zn(en)_3{}^{2+}$ may be written as

Step 1.

$$\begin{tikzcd} 2[\geqslant CeOH] \arrow[r, shift left, "\text{basic medium}"] & (\geqslant CeO^-)_3 + 2H^+ \arrow[l, shift left] \arrow[d, shift left, "Zn(en)_3^{2+}"] \\ & (\geqslant CeO^-)_2\, Zn(en)_3^{2+} + 2H^+ \arrow[u, shift left] \end{tikzcd} \qquad \text{(I)}$$

Step 2.

$$\text{(i)}\quad :NH_2-CH_2-CH_2-H_2N: + H^+ \rightarrow :H_2N-CH_2-CH_2-NH_3^+ \qquad \text{(II)}$$

$$\text{(ii)}\quad H_2\ddot{N}-CH_2-CH_2-\ddot{N}H_2 + 2H^+ \rightarrow {}^+H_3N-CH_2-CH_2-NH_3^+ \qquad \text{(III)}$$

TABLE I

Dependence of q_A and K_d value of ethylenediammines of Zn^{2+}, Ni^{2+} and Cd^{2+} on pH

Amount of the exchanger (CeO_2) = 100 mg
Concentration of en-complex ions = 0·01 M
Total volume = 20 ml.

pH	q_A values/100 mg; m-equiv.			K_d values/gm		
	Zn^{2+}	Ni^{2+}	Cd^{2+}	Zn^{2+}	Ni^{2+}	Cd^{2+}
9·0	0·100	0·008	0·010	667	41	51
9·5	0·119	0·010	0·011	847	51	56
10·0	0·139	0·012	0·012	1065	62	62
10·5	0·105	0·018	0·016	712	94	83
11·0	0·090	0·010	0·010	581	51	51

These cations (II) and (III) then start competing with the cation I; and hence the q_A value is adversely affected at higher pH.

Effect of Exchanging Ion Concentration (Table II)

The decrease in the q_A values at higher exchanging ion concentration is presumably due to the possible lowering of ionic activity at higher concentration. A diminution of exchange potential at higher concentration is well known[8].

TABLE II

Dependence of q_A values on the exchanging ion concentration

Amount of the exchanger (CeO_2) = 100 mg.
pH = 10·5, Total volume = 20 ml.

Exchanging ion concentration	q_A values/100 mg; m-equiv.		
	Zn^{2+}	Ni^{2+}	Cd^{2+}
0·01 M	0·105	0·018	0·0162
0·03 M	0·225	0·030	0·0210
0·05 M	0·176	0·036	0·0370
0·08 M	0·138	0·080	0·0085
0·10 M	0·130	0·040	0·0083

Effect of Nature of the Exchanging Ion (Tables I-IV)

The observed differences in q_A values for the different exchanging ionic species seem to be intimately connected with the charge and size of the exchanging ions[8]. Experimental observations reveal that other factors remaining the same, sorption of an ion is dependent on the ionic potential[9] (charge/radius ratio). Higher the ionic potential, greater is the sorption.

TABLE III

Dependence of selectivity on the concentration of the competing pair $Zn(en)_3^{8+} - Cd(en)_3^{2+}$

Amount of ion exchanger (CeO_2) = 100 mg;
pH = 10·5; Total volume = 20 ml.

Concentration of the competing pair		Amount sorbed per 100 mg of the solid; m-equiv.		$K = \dfrac{Zn(en)_3^{2+}}{Cd(en)_3^{2+}}$
Zn $(en)_3^{2+}$	Cd $(en)_3^{2+}$	Zn $(en)_3^{2+}$	Cd $(en)_3^{2+}$	
0·01 M	0·01 M	0·091	0·037	2·88
0·03 M	0·03 M	0·112	0·034	3·53
0·05 M	0·05 M	0·122	0·018	7·16
0·08 M	0·08 M	0·116	0·040	2·97
0·10 M	0·10 M	0·078	0·062	1·26

The en-complexes of Zn^{2+}, Ni^{2+} and Cd^{2+} are all known[10] to have octahedral symmetry and their covalent radii are[11,12]: Zn^{2+}, 1·31 Å; Cd^{2+}, 1·48 Å and Ni^{2+}, 1·39 Å. Therefore their ionic potentials will be in the order, $Zn^{2+} > Ni^{2+} > Cd^{2+}$. This order is in keeping with the observed $q_{A(max)}$ values. The fact that the selectivity quotient values follow the order of q_A values, indicate that, ionic potential is the significant factor which decides selectivity of the exchanger material.

TABLE IV

Dependence of selectivity on the concentration of the competing pair $Ni(en)_3^{2+} - Cd(en)_3^{2+}$

Amount of the exchanger = 100 mg;
pH = 10·5; Total volume = 20 ml.

Concentration of the competing pair		Amount sorbed per 100 mg of the solid; m-equiv.		$K = \frac{Ni(en)_3^{2+}}{Cd(en)_3^{2+}}$
$Ni(en)_3^{2+}$	$Cd(en)_3^{2+}$	$Cd(en)_3^{2+}$	$Ni(en)_3^{2+}$	
0·01 M	0·01 M	0·009	0·010	1·11
0·03 M	0·03 M	0·016	0·020	1·25
0·05 M	0·05 M	0·020	0·035	1·75
0·08 M	0·08 M	ppt	ppt	..
0·10 M	0·10 M	ppt	ppt	..

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1. Vesely, V. and Pekarek, V., *Talanta,* 1972, 19, 219.
2. Marinsky, J. A. and Marcus, Y., *Ion-Exchange and Solvent Extraction,* Marcell Dekker, Inc., New York, 1973, 5, 92.
3. Cotton, F. A. and Wilkinson, G., *Advanced Inorganic Chemistry,* Wiley East, Pvt. Ltd., 2nd Ed., 1970, p. 1067.
4. Vogel, A. I., *A Text-Book of Quantitative Inorganic Analysis,* ELBS, 3rd Ed., 1968, pp. 433, 435, 444.
5. Marinsky, J. A. and Marcus, Y., *Ion-Exchange and Solvent Extraction,* Marcel Dekker, Inc., New York, 1973, 5, 124.
6. Amphlett, C. B., *Inorganic Ion Exchangers,* Elsevier, 1964, p. 2.
7. Kar, K. R. and Singh, S., *Mikro. Chem. Acta,* (*Wien*), 1970, p. 279.
8. Kunin, R., *Elements of Ion Exchange,* Reinhold, New York, 1960, p. 12, 13.
9. Cartledge, G. H., *J. Amer. Chem. Soc.,* 1928, 50, 2855.
10. Heslop, L. B. and Robinson, P. L., *Inorganic Chemistry,* Elsevier, New York, Third Ed., 1967, p. 695, 756.
11. Pauling, L., *Nature of the Chemical Bond,* Cornel University Press, Thaca, New York, 1960, p. 246.
12. Heslop, L. B. and Robinson, P. L., *Inorganic Chemistry,* Elsevier New York, Third Ed., 1967, p. 123.

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LETTERS TO THE EDITOR

E 2 CONVERSION COEFFICIENTS

In general the theoretical internal conversion coefficients are supposed to be accurate. But recently Raman *et al.*[1] showed that E3 and M4 theoretical values are over-estimations by a few per cent. Hence in the present investigations a comparison of the accurate experimental results and theoretical E2 K-conversion coefficients (α_k) of Hager and Seltzer[2] is attempted to search for the possible discrepancies.

The method of Internal External Conversion (IEC) Technique for the measurement of internal conversion coefficients is standardised by Hultberg *et al.*[3] and many measurements are reported. However, the theoretical photoelectric cross sections used, are in general those of Grodstein[4] (accuracy ~5 to 15%) and Nagel *et al.*[5] (accuracy ~2%) which are not very accurate. In recent years, Scofield[6] reported very accurate theoretical photoelectric cross sections (accuracy ~0·1%). The validity of these values is established[7] Hence, most of the available E2 experimental internal conversion coefficients measured by the IEC technique[8–21] are corrected using the recent values of Scofield[6], thus improving the accuracy. To study the discrepancies between the theory and experiment, plots of [(Theory—Expt.)/Theory]% with proton number neutron number and transition energy are shown in Fig 1. In Fig. 1 the latest square fitted lines are also shown to study the variation of the discrepancy.

From a study of Fig. 1. the following conclusions can be drawn: (1) The theoretical E2 conversion coefficients are over-estimations by a few per cent and (2) the deviation between theory and experiment seems to increase with proton number, neutron number and the transition energy. These deviations may be due to the theoretical insufficiencies in the theoretical data of Hager and Seltzer[2].

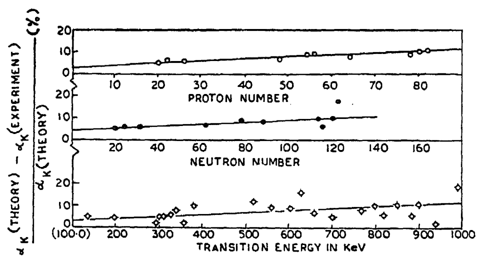

FIG. 1. Comparison between theoretical and experimental E2 conversion coefficients.

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1. Raman, S., Walkiewcz, T. A., Gunnink, R. and Martin, B., *Phys. Rev.*, 1973, **7C**, 2531.
2. Hager, R and Seltzer, E., *Nuc. Data*, 1968, **4A**, 1.
3. Hultberg, S. and Stockendal, R., *Ark. Fys.*, 1959, **14**, 565.
4. Grodstein, G W., *NBS Circular* 1957, **583**, 1.
5. Bengy Nagel, C. H., *Ark. Fys.*, 1960, **18**, 1.
6. Scofield, J. H., *Theoretical Photoionisation Cross Sections from* 1 *to* 1500 *keV*, Lawrence Livermore Laboratory, University of California, Livermore, 1973.
7. Pratt, R. H., Akavi Ron and Tseng, H. K., *Rev. Mod. Phys.*, 1973, **45**, 273.
8. Hultberg, S., Nagel, B. and Olsson Per, *Ark. Fys.*, 1961, **20**, 555.
9. —, — and —, *Ibid.*, 1968, **38**, 1.
10. Jensen, J. F. W. and Hultberg, S., *Nucl. Phys.*, 1962, **38**, 121.
11. Willard, D., *Ibid.*, 1962, **38**, 539.
12. Hankla, A. K., Hamilton, J. H. and Stockndal, R. V., *Ark Fys.*, 1963, **24**, 429.
13. Bergkvist, K. E. and Hultberg, S., *Ibid.*, 1964, **28**, 321.
14. Boyd, H. W and Hamilton, J. H., *Nucl. Phys.* 1965, **72A** 604.
15. Erman, P. and Hultberg, S., *Ark Fys.*, 1965, **30**, 101.
16. Newbolt, W. B. and Hamilton, J. H., *Nucl. Phys.*, 1964, **53**, 353.
17. El Nesr, M. Sc., and El Soyd, G. M., *Z. Phys.*, 1966, **194**, 125.
18. Zganjam, E. F., *Can. J. Phys.*, 1966, **44**, 549.
19. Raja Rao, M. and Jnanananda, S., *Ind. J. Phys.*, 1967, **41**, 55.
20. Ludington, M. A., *Nucl. Phys.*, 1968. **119A**, 398.
21. Hamilton, J. H., Antey, S. R., Van Noojien, B. and Ramayya, A. V., *Phys. Lett.*, 1966, **19**, 682.

STRUCTURAL STUDY OF $ZnNbCuO_4$

During the course of a of detailed study on ternary ($XYZO_4$ spinels) oxides[1], we could find that hardly any work is carried out on niobium-spinels, except $Zn_{2\cdot33}Nb_{0\cdot67}O_4$[2] and $ZnLiNbO_4$[3]. Romeijn[4] has studied the spinel structure from geometrical considerations which lead to a condition for formation of oxidic spinel, *i.e.*, the cationic radii should lie between 0·45 Å to 0·96 Å. The ionic radius[5] of Nb^{4+} and Nb^{5+} is 0·74 Å and 0·70 Å respectively, As the ionic radius of niobium lies well within the spinel formation region, we thought it interesting to study $ZnNbCuO_4$. The compound has been prepared for the first time by intimately mixing together the reacting oxides of A.R. grade in proper molar ratio, under acetone. The mixture was heated in a platinum boat in air in an electric furnace at 900° C for about 70 hours. The sample was cooled in the furnace. The formation of the compound was checked by X-ray diffraction patterns taken on Debye-Scherrer camera of 114·6 mm diameter, using filtered copper radiation. The pattern indicated a single phase and absence of ines due to the reacting oxides.

The crystallographic results are included in Table I. All the observed reflections are indexed for orthorhombic unit cell with dimensions $a = 9·006$ Å; $b = 8·619$ Å and $c = 9·253$ Å. It is evident from the observed reflections that the compound crystallises in a face-centered Bravais lattice.

TABLE I

Crystallographic data of $ZnNbCuO_4$

d(observed) in Å	d(calculated) in Å	$h\ k\ l$
3·231	3·228	2 2 0
2·737	2·712	3 1 1
2·583	2·583	2 2 2
2·313	2·313	0 4 0
2·117	2·154	0 0 4
2·026	2·024	4 2 0
1·868	1·857	2 4 2
1·759	1·774	1 5 1
1·674	1·665	1 1 5
1·599	1·613	4 4 0
1·515	1·516	6 0 0
1·440	1·434	0 0 6
1·369	1·367	5 3 3
$a = 9·006$ Å;	$b = 8·619$ Å	$c = 9·253$ Å

It is clear from the observed orthorhombic symmetry and the existence of planes like (420), (600), etc., that the compound is not a spinel. Though the ionic radius of Nb^{5+} is suitable for the formation of a spinel structure, large difference of ionic radii (0·70 ~ 0·96 Å) and charges of Nb^{5+} and Cu^{1+} might have been probably responsible for the instability for spinel structure.

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1. Deshpande, P. D., Kulkarni, D. K. and Khandekar, P. V., *Curr. Sci.*, 1974, **43** (15), 474.
2. Harrison, R. W. and Delgrosso, E. J., *Electrochem. Soc.*, 1963, **110**, 205.
3. Haas, *J.*, *Phys. Chem. Solids*, 1965, **26**, 1225.
4. Romeijn, F. C., *Philips Res. Reps.*, 1953, **8**, 304,
5. Sanderson, R. T., *Inorganic Chemistry*, . New York; Reinhold, 1966.

METHOD OF DETERMINING AN INDEX OF THE INFLUENCE OF VIBRATION-ROTATION INTERACTION ON FRANCK-CONDON FACTORS IN 2-ATOM MOLECULES

IN the study of intensities of electronic spectra of diatomic molecules, it is usual practice to ignore the effect of vibration-rotation interaction[1] in the calculation of Franck-Condon (FC) factors. Learner and Gaydon[2] were the first to show the error resulting from the neglect of centrifugal distortion of the potential curve in the case of OH. Since the relative intensity distribution in the diatomic molecular band system is understood in terms of FC factors, the dependence of these quantities on the rotational quantum number J has been studied by several investigators[2-10] and is shown to be significant for some molecular transitions. It is the purpose of this note to report a simple method for understanding the dependence of FC factors on J.

The effective potential for a vibrating rotator has a minimum given by

$$r_0 = r_e\,[1 + 4\,B_e{}^2\,J\,(J + 1)/\omega_e{}^2] \qquad (1)$$

which is more nearly tde equilibrium internuclear distance for the rotating molecule on the basis of the Morse-Pekeris model[11] with slight modifications[7] where r_e, B_e and ω_e are the usual spectroscopic constants. Thus a principal effect of vibration-rotation interaction[5] is the displacement of the radial co-ordinate of the minimum to larger r_0 values with increasing J. The FC factor depends on Δr_e ($= r_e' \sim r_e''$), the separation between the minima of the potential energy curves[12-14] for the two participating electronic states. Similarly FC factor $(q_{v''J''}^{v'J'})$ depends on Δr_0 ($= r_0' \sim r_0''$) in the present case. The difference ($\Delta r_0 - \Delta r_e$) could serve as an index for the influence of vibration-rotation interaction on FC factors.

The values of r_0 for different J have been calculated using the expression (1) for the various electronic states of diatomic molecules. In the present investigation, the results of Δr_0 and $(\Delta r_0 - \Delta r_e)$ are given in Table I for molecular band systems of astrophysical importance along with Δr_e. In the same table reported $(q^{v'J'}_{v''J''})$ for N_2 (B–A), OH (A–X) and CN (B–X) systems[9,10] have been entered for (0, 0) band.

TABLE I

Band system	J	Δr_0 Å	Δr_e Å	$q^{v'J'}_{v''J''}$	$(\Delta r_0 - \Delta r_e)$ Å
OH (A–X)	2½		0·0415	0·907	0·055
	50½	0·0965		0·291	
N_2 (B–A)	0		0·0740	0·339	0·008
	100	0·0818		0·312	
*CN (B–X)	0		0·0212	0·920	0·001
	100	0·0226		0·928	
AlH (A–X)	0		0·0001	..	0·087
	25	0·0874		..	
MgH (A–X)	0		0·0510	..	0·042
	100	0·0934		..	
CH (A–X)	0		0·0183	..	0·001
	100	0·0190		..	
C_2 (Swan)	0		0·0459	..	0·003
	100	0·0492		..	

* Molecular constants from Herzberg[1]; for the rest from Rosen[15].

It is clear from the results in the table that FC factors are not affected significantly with increase of J for N_2 (B–A), CN (B–X) band systems. For these systems, the value of $(\Delta r_0 - \Delta r_e) < 10^{-2}$ Å. On the other hand OH (A–X) system shows a large variation in the FC factors with increase of J reflecting the influence of vibration-rotation interaction. For this system, the value of $(\Delta r_0 - \Delta r_e)$ is large compared to those of the other band systems. Thus it is observed that larger the value of $(\Delta r_0 - \Delta r_e)$ greater will be the influence of vibration-rotation interaction. In the context of these observations, it can be predicted that the vibration-rotation interaction may influence the FC factors significantly for AlH (A–X) and MgH (A–X) band systems for which $(\Delta r_0 - \Delta r_e)$ values are comparable with that of OH. We urge those engaged in the study of intensities of these band systems of astrophysical importance to compute FC factors including the vibration-rotation interaction. We can further infer that the influence of this interaction is negligible in the case of CH (A–X) and C_2 (Swan) band systems as $(\Delta r_0 - \Delta r_e)$ value is small. In such cases laborious calculation of FC factors with vibration-rotation interaction need not be computed. Hence the knowledge of $(\Delta r_0 - \Delta r_e)$ will be useful before undertaking tedious calculation of FC factors involving vibration-rotation interaction. The details of this method along with the results of other band systems will be published elsewhere.

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1. Herzberg, G., *Molecular Spectra and Molecular Structure. Spectra of Diatomic Molecules,* (Second Edn.), D. Van Nostrand, New York, 1950.
2. Learner, R. C. M. and Gaydon, A. G., *Nature,* 1959, 183, 242.
3. —, *Proc. Roy. Soc.* (London), 1962, 269 A, 311.
4. Jain, D. C. and Sahni, R. C., *Proc. Phys. Soc.* (London), 1966, 88, 495.
5. Villarejo, D., *J. Chem. Phys.,* 1968, 49, 2523.
6. —, Stockbauer, R. and Inghram, M. G., *Ibid.,* 1969, 50, 1754.
7. Shumaker, J. B., *J. quant. Spectroc. Radiat. Transfer.,* 1969, 9, 153.
8. Generosa, J. I. and Harris, R. A., *J. Chem. Phys.,* 1970, 53, 3147.

9. Chakraborty, B., Pan, Y. K. and Chang, T. Y., *Ibid.*, 1971, 55, 5147.
10. — and —, *Ibid.*, 1972, 56, 3722.
11. Pekeris, C. L., *Phys. Rev.*, 1934, 45, 98.
12. Tawde, N. R., Patil, B. S., Murthy, N. S. and Katti, M. R., *Physica*, 1957, 23, 154.
13. —, and Murthy, N. S., *Ind. J. Phys.*, 1957, 31, 391.
14. Murthy, N. S. and Murthy, B. N., *Nature*, 1969, 223, 181.
15. Rosen, B., *Spectroscopic Data Relative to Diatomic Molecules*, Pergamon Press, Oxford, 1970.

AMINO ACID COMPOSITION C WILD LEGUMES

PRELIMINARY investigation revealed the presence of 23·2%, 25·3% of proteins in the seeds of *Cassia tora* and *Cesalpinia bounducella* respectively. In view of their high protein content, the nutritional characteristics of their protein isolates are assessed.

The mature seeds were coarsely ground, defatted with petroleum ether (B.P. 60–80° C) and the seed was repeatedly extracted with cold alkaline 10% brine for several hours and filtered. The filtrate on acidification with 10% acetic acid precipitated the proteins in yields of 15·6% and 17·4% from *Cassia tora* and *Cesalpinia bonducella* seeds respectively. Protein isolates were hydrolysed to negative biuret test by refluxing with a mixture of 6N.HCl and 80% formic acid taken on 1 : 1 ratio by volume. After humin removal, the protein hydrolysate was evaporated to a syrupy mass, which was vacuum desiccated. The mass was then dissolved in 10% isopropanol and was qualitatively analysed by various paper chromatographic techniques[1-5] using different solvents and different staining reagents[6-9]. The quantitative determinations of amino acids were made by photometric ninhydrin method[10]. The results are shown in Table I. The addition of 80% formic acid to 6 N.HCl during hydrolysis of the protein was found to be advantageous in two ways : (1) It reduced the time required for complete hydrolysis of the protein. A period of 12–16 hours refluxing was needed with this mixed acid solution, as against 18–24 hours required with 6 N.HCl. (2) It reduced amino acid losses, possibly by checking the oxidative type of degradation of some of the amino acids, particularly lysine, threonine, methionine and tryptophane.

Cesalpinia bonducella seeds are found to be rich in all essential amino acids particularly lysine methionine, threonine, leucine and isoleucine. These seeds are, however, dificient in tryptophane. *Cassia tora* seeds appear to be rich in all essential amino acids particularly, methionine and tryptophane but are deficient in lysine.

TABLE I

Amino acid composition of protein isolates of wild legumes

(Amino acid percentage expressed in g/16 g of nitrogen)

Sl. No.	Per cent of Amino acid	*Cesalpinia bonducella*	*Cassia tora*
	Indispensable		
1.	Lysine	6·83	3·70
2.	Histidine	3·40	2·42
3.	Threonine	8·2	9·2
4.	Phenylalanine	5·22	4·80
5.	Valine	8·5	6·4
6.	Methionine	0·9	1·6
7.	Tryptophane	0·4	0 9
8.	Leucine and isoleucine	15·4	15·1
	Semi-Indispensable		
9.	Serine*	7·0	7·4
10.	Glycine (indispensable for chicks)	9·2	10·2
11.	Tyrosine**	0·8	3·86
12.	Cystine†	0·9	0·8
13.	Arginine (indispensable both for rats and chicks)	0·2	6·6
	Dispensable		
14.	Glutamic acid	9·0	10·6
15.	Aspartic acid	8·4	6·8
16.	Alanine	8·6	8·4
1..	Proline‡	0·2	0·6
18.	γ-Amino butyric acid	Traces	Traces

Mean values of 10 determinations

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* Serine exerts sparing effect upon glycine.
** Tyrosine exerts sparing effect upon phenylalanine.
† Cystine exerts sparing effect upon methionine.
‡ Optical density measured at 440 nm.

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1. Matthias, W., *Naturwissenschaften*, 1954, 41, 17.
2. Giri, K. V., *J. Ind. Inst. Sci.*, 1953, 35 (2), 149.
3. — and Rao, N. A. N., *Ibid.*, 1953, 35 (4), 343.

4. Martin, A. J. P., *Ann. Rev. Biochem.*, 1950, 19, 517.
5. Joshi, S. S., *Ph.D. Thesis.* Ravishankar University, 1974, p. 75.
6. Muting, D., *Naturwissenschaften,* 1952. 39, 303.
7. Sakaguchi, S. S., *J. Biochem.* (Tokyo), 1925, 5, 25.
8. Saifer, A. and Oreskes, I., *Science,* 1954, 119, 124.
9. Jepson, J. B. and Smith, I., *Nature,* 1953, 172, 1100.
10. Dent, C. E., *Biochem. J.*, 1947, 41, 240 ; 1950, 47, 223.

KINETICS OF SUBSTITUTION OF POTASSIUM TRIS (OXALATO) CHROMATE (III) BY ORTHOPHOSPHORIC ACID

THE complexation of hexaaquochromium (III) ions by phosphoric acid has been reported recently by Lahiri[1]. The rate of substitution of tris (oxalato) chromium (III) ion by phosphoric acid which we report in the present communication is found to be relatively faster, and is dependent on the molarity of phosphoric acid as well as the concentration of the metal complex.

Potassium tris (oxalato) chromate (III) was prepared[2] and its purity confirmed by analysis[3]. All other chemicals used were of analytical grade. Ionic strength was kept at 3·0 with respect to sodium nitrate. The rate of substitution was followed with a Unicam SP 600 spectrophotometer at 420 nm, by following the reactant, tris (oxalato) chromium (III), which has got an absorption maximum at that wavelength.

The order with respect to time $(n_t = 2{\cdot}80)$ was found to be greater than the order with respect to concentration $(n_c = 1{\cdot}08)$ indicating inhibition of the reaction by the product[4]. Hence initial rate methods were employed to evaluate the rate constants by the method described by Laidler[4]. The order of the reaction with respect to the metal complex and orthophosphoric acid was found to be 1 and 2 respectively. A plot of initial rate *vs* $[H_3PO_4]^2$ gives a straight line with a positive intercept on the rate axis showing that the reaction goes along two paths : one with a rate directly proportional to $[H_3PO_4]^2$ and the other with a rate independent of $[H_3PO_4]$. Increasing ionic strength increases the rate constant.

Ion exchange studies with the green product species failed to show the presence of either cationic or anionic phosphato complexes. This suggests that the product species may be neutral probably of the type $[Cr(H_2PO_4)(HPO_4)]$[5]. The spectrum of the product gave a maximum at 440 nm.

The complexation of $Cr(H_2O)_6{}^{3+}$ by H_3PO_4 was also carried out. The product species, green in colour (which was also found to be neither cationic nor anionic by ion exchange studies) in this case gave identical absorption spectrum with an absorption maximum at 440 nm. To evaluate the rate constants the absorption of the product at 440 nm was followed. Results are presented (Table I).

TABLE I

Temperature = 28·5° C Ionic strength = 3·0

$Cr(ox)_3{}^{3-}$ (M)	H_3PO_4 (M)	10^2 Initial Rate moles lit^{-1} min.$^{-1}$	Pseudo first order rate constant, k min.$^{-1}$
0·002	3·0	0·44	2·20
0·003	3·0	0·68	2·26
0·004	3·0	0·90	2·25
0·005	3·0	1·14	2·28
0·006	3·0	1·34	2·23*
0·004	3·0	0·32*	0·80
0·004	3·0	0·59**	1·47**
0·004	1·0	0·38	0·95
0·004	1·5	0·46	1·15
0·004	2·0	0·58	1·45
0·004	2·5	0·72	1·80
0·004	3·5	1·11	2·77
0·004	4·0	1·36	3·40

$Cr(H_2O)_6{}^{3+}$ (M)	H_3PO_4 (M)	10^2 Initial rate moles lit^{-1} min.$^{-1}$	Pseudo first order rate constant, k min.$^{-1}$
0·016	2·0	0·92	0·58
0·016	2·5	0·96	0·60
0·016	3·0	1·03	0·64
0·016	3·5	1·16	0·72
0·016	4·0	1·30	0·81

* $\mu = 1{\cdot}0$.
** $\mu = 2{\cdot}0$.

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1. Lahiri, S. C., *J. Indian Chem. Soc.*, 1975, 52, 478.
2. Brauer, G., *Hand Book of Preparative Inorganic Chemistry,* 2nd. Ed., Academic Press, N.Y., 1965, Vol. 2.
3. Krishnamurti, K. V. and Harris, G. M., *J. Phys. Chem.*, 1960, 64, 346.
4. Keith, J. Laidler., *Chemical Kinetics,* Ch. I, McGraw-Hill, 2nd Ed., 1965, p. 16.
5. Jameson, R. F. and Salmon, J. E., *J. Chem. Soc.*, 1955, p. 360.

MAGNETIC PROPERTIES OF 4-AMINOANTIPYRINE COMPLEXES OF SOME LANTHANIDE PERCHLORATES AND NITRATES

RECENTLY we have prepared and characterized some complexes of 4-aminoantipyrine (aap) with perchlorates[1] and nitrates[2] of La (III), Pr (III), Nd (III), Sm (III), Gd (III), Dy (III) and Y (III). In view of the fact that comparatively little data is available in the literature on the magnetic properties of lanthanide complexes, it was considered worthwhile to study the magnetic behaviour of these new complexes and to compare the values with (*a*) theoretical predictions on the basis of complete spin-orbit coupling[3] and (*b*) the Van Vleck values[4]. The results of such a study are presented in this communication.

Materials and Methods

The following 14 complexes were studied, with general formulae: $[Ln(aap)_6](ClO_4)_3$ and $[Ln(aap)_3(NO_3)_3]$ where Ln = La (III), Pr (III), Nd (III), Sm (III), Gd (III), Dy (III) and Y (III). These complexes were prepared and analysed by procedures described earlier[1,2]. The magnetic susceptibilities were determined by the Guoy method at ambient temperatured (28± 2° C).

The measured susceptibilities were corrected for the diamagnetism of the rest of the molecule using Pascal's constants[5]; a correction for the diamagnetism due to the completely filled orbitals of the lanthanide ion was also applied. The moments were calculated using the Curie law formula $\mu_{eff} = 2 \cdot 84 \sqrt{\chi'_M - T}$. It may be mentioned here that these complexes represent magnetically dilute systems and therefore the Weiss constant can be neglected[6]. The experimentally determined magnetic moments are presented in Table I.

Discussion

Theoretical magnetic moments were calculated using the formula $\mu = g\sqrt{J(J+1)}$ which is valid when appreciable spin-orbit coupling exists, as is the case in the lanthanide systems. The '*g*' factor here was calculated from the relation

$$g = 1 + \frac{J(J+1) + S(S+1) - L(L+1)}{2J(J+1)}$$

TABLE I

Magnetic properties of some lanthanide complexes

Complex	χ'_M corrected $\times 10^{-6}$	μ experimental* B.M.	μ theoretical** B.M.	μ theoretical† B.M.
$[La(aap)_6](ClO_4)_3$	10	0	0	0
$[Pr(aap)_6](ClO_4)_3$	4419	3·26	3·58	3·62
$[Nd(aap)_6](ClO_4)_3$	5346	3·58	3·62	3·68
$[Sm(aap)_6](ClO_4)_3$	1076	1·61	0·85	1·55–1·65
$[Gd(aap)_6](ClO_4)_3$	25331	7·80	7·94	7·94
$[Dy(aap)_6](ClO_4)_3$	50614	11·03	10·64	10·60
$[Y(aap)_6](ClO_4)_3$	12	0	0	0
$[La(aap_2(NO_3)_3]$	17	0	0	0
$[Pr(aap)_3(NO_3)_3]$	5243	3·55	3·58	3·62
$[Nd(aap)_3(NO_3)_3]$	6263	3·88	3·62	3·68
$[Sm(aap)_3(NO_3)_3]$	976	1·58	0·85	1·55–1·65
$[Gd(aap)_3(NO_3)_3]$	30584	8·57	7·94	7·94
$[Dy(aap)_3(NO_3)_3]$	50801	11·07	10·64	10·60
$[Y(aap)_3(NO_3)_3]$	39	0	0	0

* using the Curie formula.
** using the simple spin-orbit coupling model,
† using the Van Vleck values.

Although this g value is applicable only to gaseous ions, it may be presumed that in view of the extensive shielding of the 4f orbitals, the lanthanide ions in ligand fields behave as gaseous ions. Besides, these theoretical values calculated on the basis of the simple spin-orbit coupling model, the values obtained by Van Vleck and Frank from more refined calculations[4] are also included in Table I for comparison.

La and Y complexes are diamagnetic, as is to be expected. It may be seen from Table I that the experimental moments compare favourably with the simple-model theoretical values except in the case of Sm. In the case of Sm, the experimental moment is almost double the theoretical value. The reason for this is that in Sm, the first excited J state is sufficiently close to the ground state, causing an increase in the magnetic moment. Such an effect is observed in the case of both the perchlorate and nitrate complexes of Sm. It may be noted that the Van Vleck values agree closely with the experimental values in all cases (including Sm)

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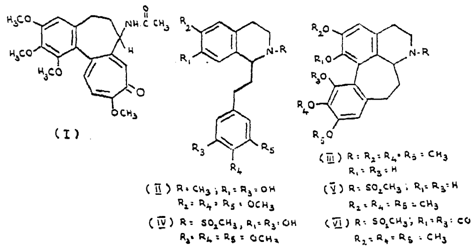
1. Jacob Chacko, Prabhakaran, C, P. and Nair, C. G. R., *Indian J. Chem.*, 1975, **13**₂ 411.
2. — and —, *J. Inorg. Nucl. Chem.* (in press).
3. Drago, R S., *Physical Methods in Inorganic Chemistry*, Affiliated Eastwest Press, New Delhi, 1971.
4. Van Vleck, J. H. and Frank, N., *Phys. Rev.*, 1929, **34**, 1494.
5. Selwood, P. W., *Magnetochemistry*, Interscience, New York, (1960).
6. Moeller, T. and Horowitz, E. P., *J. Inorg. Nucl. Chem.*, 1959, **12**₂ 49.

PHENOLIC OXIDATION OF A 1-PHNETHYL-ISOQUINOLINE

Colchicine (1), the alkaloid of colchicum autumnale, is of considerable use in medicine particularly as abortifacient[1]. Its usefulness for gout and skin carcinoma has also been referred[2].

The observation that autumnaline (II) transformed biogenetically into colchicine[3] (1) prompted the study of phenolic oxidation as a biogenetic mode of synthesis towards analogues of colchicine (1). Potassium ferricyanide oxidation of autumnaline (II) gave (III) by ortho-ortho coupling[4]. Manganese dioxide in chloroform as oxidizing agent has been considered to have steric preference for para-coupling compared to ortho-coupling. Franck pointed out that phenolic oxidation of the compound absorbed on silica gel would be similar in process to enzyme reaction in the plant cell[5].



In a phenolicox idation study of 1-(3′-Hydroxy-4, 5′-dimethoxyphenyl)-6 methoxy-7-hydroxy-N-mesyl-1, 2, 3, 4-tetrahydroisoquinoline (IV), using manganese dioxide in chloroform, a product has been obtained. Analysis of the compound is in agreement with the molecular formula $C_{21}H_{25}O_7$ NS and gave the I.R. absorption at 3420, 2920, 1600 δ 1140 cm^{-1}. Ultraviolet spectrum with absorptions at 242 nm, 263 nm and 298 nm showed it to be a homoaporphine type compound[6].

NMR spectrum of the compound recorded as 2·71 δ (3H, s, SO_2CH_3); 3·81 δ (3H, s —OCH_3) 3·88 δ (3H, s, —OCH_3); 3·90 δ (3H, s —OCH_3); 4·42 δ (1H, m. tert H); 6·58 δ (1H, s, aromat H); 6·79 δ (1H, s—aromat H); 7·51 δ (2H, m, hydroxyl H) is in agreement with the structure (V) for the product.

Mass spectrum recorded at 70 ev registers a peak at 418 ($M^+ - 17$) and a high resolution mass spectrum gave a base peak at 435 (M^+) and a subsequent peak at 418 (95%, $M^+ - 17$). Further to establish the presence of two hydroxyl groups in the compound mass spectrum of the acetate (VI) has been studied, which reads as 519 (24, M^+); 477 (58, $M^+ - CH_2CO$) and 435 (100, $M^+ - 2.CH_2CO$).

A study of the mass spectrum of the acetate (VI) indicated the presence of two phenolic hydroxyl groups in the compound. The observation of ($M^+ - 17$) as a base peak in the mass spectrum of the compound is in agreement with the observation made by Jackson and Martin in the mass spectra of the aporphin alkoloids[7]. Hence the structure of the oxidation product is established as (V). Further study of the oxidation of the compound (IV) under various conditions is under progress.

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1. Watt, J. M. and Breyer-Brandwijk, M. G., *The Medicinal and Poisonous Plants of Southern and Eastern Africa*, Livingstone, Edinburgh and London, 1962.
2. Roswitha Schmidt, *Univsrsitas* (Engl. Edit.). 1974, **16**, 261.
3. Barker, A. C., Battersby, A. R., Mcdonald, E. Ramage, R. and Clements, J. H., *Chem. Comm.*, 1967, p. 390.
4. Battersby, A. R , Bohler, P., Munro, M. G. H. and Ramage, R., *Ibid.*, 1969, p. 1066.
5. Franck, B., Dunkelmann, G. and Lubs H. J., *Angew. Chem.*, 1967, **79**, 1066.
   —, — and —, *Ibid.*, 1967, **79**, 987.
6. Battersby, A. R., Bradbury, R. B., Herbert, R. B., Munro, M. G. H. and Ramage, R., *Chem. Comm.*, 1967, p. 450.
7. Jackson, A. H. and Martin, J. A., *Ibid.*, 1966, p. 2181.

POTASSIUM-ARGON AGES OF MICAS FROM THE PEGMATITES OF BHILWARA, RAJASTHAN, INDIA

THE pegmatites that occur in the three major Precambrian rock groups of Rajasthan, namely, the Banded Gneissic Complex, the Aravalli Formation and the Delhi Formation, have generally been related with the late intrusive phases of the Aravalli and Delhi Orogenic cycles. Heron[1] and Gupta[2] assigned them pre-Delhi (pre-Aravalli) and post-Delhi ages whereas Crookshank[3] asd Sharma[4] regarded them as of post-Aravalli (pre-Delhi) and post-Delhi ages, respectively. According to Heron[1], the medium grained, foliated pre-Delhi pegmatites, characterised by the absence of tourmaline, beryl and radioactive minerals and scarcity of muscovite, were disturbed by later movements. The post-Delhi pegmatites were described as coarse grained, unfoliated and undisturbed rocks containing these minerals. However, these post-Delhi pegmatites are more widespread in the older Aravalli Formation and the Banded Gneissic Complex than in the Delhi Formation.

Absolute age determinations on Rajasthan pegmatites have been attempted earlier by Pb-U[5-7], K-Ar[7], Rb-Sr[8] and fission track[9-12] methods and, excepting one 1935 Ma. Rb-Sr age of a pegmatite in the Gneissic Complex[8], all the reported dates are less than 1350 Ma.

We have determined K-Ar dates on four samples of muscovite and lepidolite from Bhunas and Potlan pegmatities of Bhilwara District, Rajasthan. The pegmatite of Danta Mine, Bhunas, is an oval, multi-quartz core, asymmetrically zoned, concordant intrusive body in the Aravalli garnet mica schist; it contains workable lodes of muscovite, giant crystals of beryl and tourmaline, cytolite, uraninite and replacement pods of cleavelandite[13]. The pegmatite of Krishna Mine, Potlan, is a very irregular body exhibiting both concordant and discordant relationship with the host Aravalli garnet-mica schist. Stained muscovite and black tourmaline occur in the border and wall zones. Lepidolite and rebellite occur in the cleavelandite replacement bodies.

Potassium was determined on EEL flame photometer; the results quoted in Table I are the averages of three determinations on each sample. Argon was determined on a modified A.E.I. MS 10 mass-spectrometer[14], using ^{38}Ar as spike. The average of the two muscovite dates from Bhunas pegmatite is 1024 ± 40 Ma. The average of the two lepidolite and muscovite dates from Potlan pegmatite is 848 ± 35 Ma., *i.e.*, about 175 Ma. younger than that for the Bhunas pegmatite. These results compare well with those of Vinogradov *et al.*[7], who determined 1100 ± 50 Ma. age for Bhivarla (Bhil-

TABLE I

K–Ar dates of Bhilwara pegmatites, Rajasthan (India)

Sample No.	Mineral	Location	% K	Vol. of ^{40}Ar radiogenic sec/gm $\times 10^{-4}$	% ^{40}Ar radiogenic	Age in million years*
BH/78	Muscovite	Danta Mine Pegmatite, Bhunas (32 km SW of Bhilwara)	8·70	4·8275	97·3	1042±42
BH/99	Muscovite	do.	8·66	4·5931	96·9	1006±40
PT/5	Lepidolite	Krishna Mine Pegmatite, Potlan (50 km SW of Bhilwara)	9·51	3·8488	97·8	811±32
PT/6	Muscovite	do.	8·83	3·9830	97·7	885±35

*Constants used: $\lambda_e = 0{\cdot}584 \times 10^{-10}\ \text{yr}^{-1}$, $\lambda_\beta = 4{\cdot}72 \times 10^{-10}\ \text{yr}^{-1}$, ^{40}K/K (atomic) = 0·0119%, wara ?) pegmatite, using 1080 Ma. ^{207}Pb/^{206}Pb age of cyrtolite and 1040 Ma. K-Ar age of muscivite, and 900 ± 50 Ma. K-Ar age for Ajmer and Rajgarh pegmatites, respectively.

The Banded Gneissic Complex, which forms the basement of the Aravalli rocks, represents part of an old craton intruded by granites as old as 2,500 Ma.[8,15]. Based on K–Ar determinations, Sarkar *et al.*[15] suggested that the Aravalli Orogenic cycle closed between 1500 Ma. and 950 Ma. and the Delhi cycle closed between 860 Ma. and 734 Ma., the latter conforming to the views of Holmes *et al.*[5]. Rb–Sr age determinations, however, reveal that folding and uplift of the Aravalli rocks was accompanied by granitic intrusions at about 2100–1900 Ma. and the folding and uplift of the Delhi rocks was accompanied by granite and pegmatite intrusions at 1700–1650 Ma., 1400 Ma., 1200 Ma. and 1000–950 Ma. which continued in a pulsating manner resulting into pegmatites dated at around 850, 750, 650 and 580 Ma.[8].

The Bhunas and Potlan pegmatites, therefore, appear to be post-Delhi in age, though they occur in the Aravalli schists. The fact that most of the determinations have given dates between 750 and 1100 Ma. indicates that this was the most 'active' period of pegmatite activity in Rajasthan and the still lower ages probably indicate sporadic intrusive phases, cooling or subsequent events of mineralisation mainly of hydrothermal character.

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---

1. Heron, A. M., *Mem. Geol. Surv. India,* 1953, **79**, 389.
2. Gupta, B. C., *Ibid.,* 1934, **65** (2), 107.
3. Crookshank, H., *Trans. Min. Geol. Met. Inst. India,* 1948, **42**, 105.
4. Sharma, N. L., *Press. Add., Geol. Geog. Section, 40th Ind. Sci. Congr.,* 1953, p. 27.
5. Holmes, A., Smales, A. A., Leland, W. T. and Nier, A. O., *Geol. Mag.,* 1949, **86**, 288.
6. Aswathanarayana, U., *Bull. Geol. Soc. Amer.,* 1959, **70**, 111.
7. Vinogradov, A., Tugarinov, A., Zhykov, C., Stapnikovt, N., Bibikova, E. and Khorre, K., *Rep. 22nd Intern. Geol. Congr.,* 1964, **10**, 553.
8. Crawford, A. R., *Canad. J. Earth Sci.,* 1970, **7**, 91.
9. Mehta, P. P. and Nagpaul, K. K., *Ind. J. Pure and Appl. Phys.,* 1970, **8**, 397.
10. Mehta, P. P. and Nagpaul, K. K., *Pure and Appl. Geophys.,* 1971, **87**, 174.
11. Gupta, M. L., Nagpaul, K. K. and Mehta, P. P., *Canad. J. Earth Sci.,* 1971, **8**, 1491.
12. Nagpal, M. K. and Nagpaul, K. K., *Ibid.,* 1974, **11**, 519.
13. Mehta, P. K., *Bull. Ind. Geol. Assoc.,* 1974, **7**, 67.
14. Rex, D. C. and Dodson, M. H., *Eclogae geol. Helv.,* 1970, **63**, 275.
15. Sarkar, S. N., Polkanov, A. A., Gerling, E. K. and Chukrov, F. V., *Sci. and Cult.,* 1964, **30**, 527.

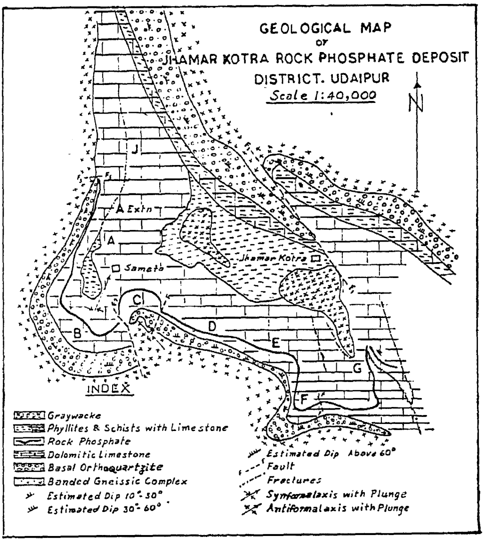
A SHORT NOTE ON STRUCTURE OF JHAMARKOTRA ROCK PHOSPHATE DEPOSIT, DISTRICT UDAIPUR, RAJASTHAN

A LARGE deposit of rock phosphate was discovered by the State Department of Mines and Geology, Rajasthan, in the year 1968 at Jhamarkotra near Udaipur. The area covered under present investigation falls between longitudes 73° 49′ to 73° 52′ and latitude 24° 27′ to 24° 29′ covering approximately 50 square kilometers. Detailed investigation by the Department of Mines and Geology has proved a reserve of 37·71 million tonnes of phosphorite of varying grades ranging from 12% to 38% P_2O_5 content.

The first record of the geology of the area is given by Heron (1953) who identified the rocks as belonging to PreCambrian age. He classified the rocks into three classes, (1) Banded and streaky gneisses grouped under the Banded Gneissic Complex, (2) Argillaceous, quartzitic and dolomitic rocks grouped under the Aravalli system, and (3) Argillaceous, arenaceous and calcareous rocks along the main Aravalli hill range grouped under Delhi system.

In recent years, several workers like P. K. Mathur (1963–64), Iqbaludin (1965–66), and G. J. Chandak (1965–66) from the G.S.I. have studied the area on a regional and detailed basis and their results have been compiled by C. S. Raja Rao, B. C. Poddar *et al.* (1971).

According to C. S. Raja Rao *et al.* (*op. cit.*), Heron's correlation of Debari conglomerate quartzite formation to the east of Udaipur with the Alwar formation of Delhi group has been found to be inconsistent with the field facts. This rock group has been considered as co-eval with the basal Aravalli rocks and the sequence at Debari is inverted, with the Aravallis structurally overlain bf the basement. As will be evident from the foregoing discussion, the authors also feel that the succession in the northern portion of Jhamarkotra area is inverted as we get phyllites at the base, followed by limestones and quartzites. Furthermore, mobilisation of basement in Jhamarkotra area may be seen along the fault F_1F_1 (Map).



Systematic mapping of the area has revealed the following succession of rocks :

Graywacke.
Phyllites and schists.
Dolomitic limestone.
Rock phosphate.
Dolomitic limestone.
Basal orthoquartzites/Arcose.
Unconformity
Banded gneissic complex.

The present paper deals with the major structural elements in the area and their relationship with the mineralization of rock phosphate. The aerial photographs of the area taken on a scale of 1 : 40,000 were studied with the help of mirror stereoscope. The interpretation of the structural studies were checked in the field. A number of readings of attitudes of beds (Slipplanes), joints and cleavage were recorded and folds analysis was carried out.

Structure :

The area forms a part of the Aravalli Synclinorium. Banded gneissic complex represents the oldest rock units exposed all around the fringe of the basin. Presence of stromatolitic structure in rock phosphate and the enclosing dolomitic limestone indicate shallow water deposition. Presence of ripple-marks and cross-beds in the basal orthoquartzites may represent the part of ancient shoreline in the area.

The Jhamarkotra synform extends for a length of about 6–7 km in NW–SE direction with a width of about 6 km. In relation with the other phosphate deposits, such as Maton, Kanpur, Bargaon, Neemuch Mata and Sisarma, it is believed that the Jhamarkotra deposit represents a separate tectonic basin.

There have been three to four generations of deformation in the Aravalli orogeny and the Jhamarkotra area has also been affected by such structural disturbances. In the tectonic history of the Aravallis, it is considered that the first compressional stress acted from the south-east direction direction which resulted in the regional strike direction trending North-each—South-west. The general trend of strike in Block A and A extension at the western part of the Jhamarkotra deposit swerves from NNE–SSW to almost East-West and WNW–ESE which is more pronounced in Blocks D and E in the central part of the deposit. The quartzite to the north of the area show NNW–SSE strike. These different attitudes of bedding planes indicate the different stresses that might have prevailed in the early tectonic phase of the area as a result of which the rocks were subjected to cross-folding.

A number of fold structures have been noted in the area. In the south-western corner of the area in which Blocks A and B are situated, there is a synform whose axis trends almost NNE–SSW with a gentle plunge to the NNE. The eastern limb of this fold has been dissected by a fault F_2F_2 trending NNW–SSE which has displaced the quartzite for about 300 m. Due to the effect of this fault, the rock phosphate band has been discontinued for about 200 m. in length. Complementary to this synform we get an antiformal structure in Block C with a gentle plunge to the NW.

Due to the resultant effect of these two fold axis the hills near the village Sameta display a dome-like structure with dips spreading in all the directions. The eastern limb of this antiform has been truncated by a minor fault F_4–F_4 just at the junction of Blocks C and D which striges NNE-SSW and has displaced the ore body. The ore body in the central part of the deposit in block D and part of Block E present a monoclinial limb. Exposures in the mine face of Block D at places show dragfolds at the contact of the rock phosphate and overlying limestone. The axis of these dragfolds point in the northern direction. Veering of strike in the south-eastern portion of the deposit in Blocks E, F and G has been noticed. In the eastern part of the area near Garhamata temple, an antiformal fold has been noticed whose limbs dip in the same direction towards East.

A major thrust fault F_1–F_1 is noticed in the north-eastern part of the deposit near the village Jhamarkotra which has displaced the quartzite ridge for about 1500 m. Due to the effect of this major thrust, a number of sympathetic fractures and faults have developed in the north-eastern portion of the area. A number of minor folds and faults are also observed in the western portion in Block A Extension and Block J. A fault F_3F_3 striking almost East-West has displaced the quartzite. The presence of cataclastic breccia in the vicinity of this fault area also confirms the same. Minor folds are noticed in the road cutting of A Extention block plunging towards west.

Structural Control of Mineralization :

The rock phosphate deposit of Jhamarkotra is synsedimentary deposit of mainly biogenic origin. The deposit shows lateral variation in the grade from one end to the other. This variation in the grade may be explained on the basis of structural events described above. Blocks D and E lying in the central part of the deposit have suffered maximum effect of compressional stresses, which have resulted in high grade ore zone from which soluble carbonates have been subsequently leached out. Again in Block B, the high grade zone represents the central part (core of the synform) while low grade ore is seen on the flanks. Due to the presence of an antiform in Block C, tensional stresses may have stretched the ore body resulting in a low grade deposit in that area. Just at the junction of Blocks D and E in the eastern saddle portion, the low grade zone may be due to tensional stresses.

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---

1. Raja Rao, C. S., "Precambrian Startigraphy of Rajasthan—A review," *Rec. Geol. Surv. Ind.*, 1971, 101, Part 2.
2. Heron, A. M., "The Geology of Central Rajputana," *Mem. Geol. Surv. Ind.*, 1953, 79, Part 1.

IRREGULAR MICROSTRUCTURES IN GODAVARI COALS

PETROGRAPHIC study of Godavari valley coals has revealed that these microconsituents with outer-edge serrated and inner-edge either concave or straight are bodies of resin. The chain-like structure is developed by concentration and fusion of resins. The reflectance of these microconstituents has been compared with that of the associated vitrinite, fusinite and fusinized resins and it is concluded that they represent similar conditions of transformation. During the microscopic study of Godavari valley coals, some peculiar features were frequently noticed in them, and an attempt has been made to determine their exact nature and also to assign them to a particular maceral group.

Discussion :

Ganju and Pant (1963) illustrated occurrence of such microconstituents from Raniganj coals and described them as resin globules arranged in a chain-like fashion which appear smooth and without borders. From their arrangement it appears that they have been concentrated due to the deposition of a sporangium body which is generally protected by waxy, resinous gummy covering. Ganju (1958) from Rampur coals and Ramana Rao (1968) from Kothagudium coalfields have described similar structures as thick cuticles with one edge serrated. In the present study of Godavari valley coals, a number of similar microconstituents in different states of preservation have been recorded. They usually show one edge as serrated and the other as either straight or slightly concave and they are grouped together in a chain-like form. The reflectance of these constituents is equal to that of either the associated vitrinite or fusinite. In some cases, it is inbetween the two.

Figure 1 shows the reflectance of these microconstituents to be equal to that of the associated

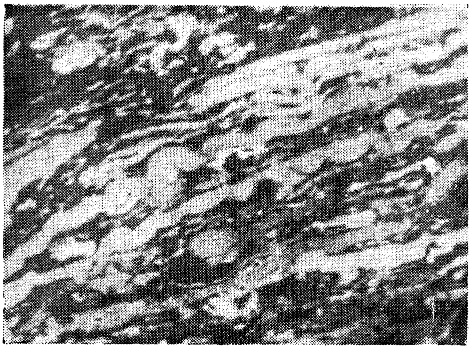

FIG. 1.

vitrinite, while Figs. 3 and 4 show the reflectance as equal to that of the associated fusinite. Figure 2 shows that the reflectance is inbetween the two. These microconstituents, in most of the cases observed, appear as rounded or oval-shaped bodies with or without a cavity at the centre and they seem to have been fused together to form a chain-like structure (Figs. 3 and 4). It appears that the outer rim breaks at the point of fusion, as seen in Fig. 4. When all the cavities are interconnected they produce a peculiar structure which appears serrated on the outer-edge and straight to concave on the inner-edge. An exactly similar structure is presented by rounded sponge type resins with a thick outer margin, grouped together in the form of chain and in a fused condition (Fig. 4). Some of the resins are broken at the points of fusion or attachment and so present a convex outer-edge and a concave inner-edge.

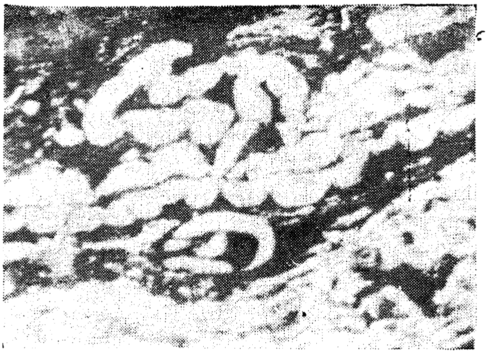

FIG. 2.

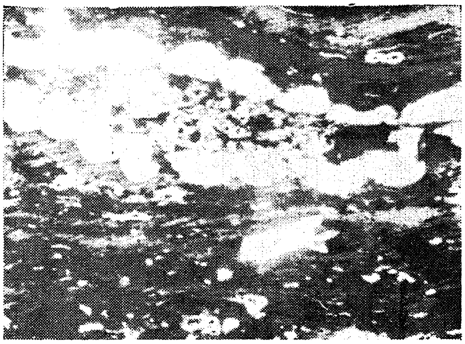

FIG. 3.

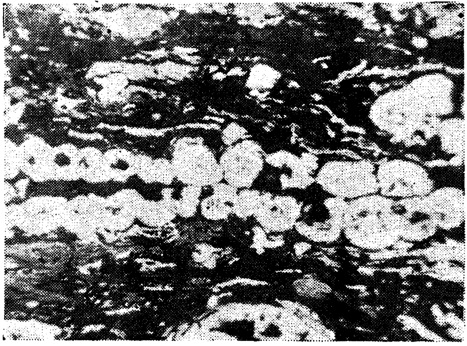

FIG. 4.

The close similarity of these microconstituents with the associated resin bodies and their appearance similar to simple and fusinized resins through an intermediate stage (Ramana Rao and Moiz), suggest that they are resinous bodies fused together which have undergone transformations similar to those that produced vitrinite, semi-fusinite and fusinite in Godavari valley coals.

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Hyderabad, *October* 27, 1975.

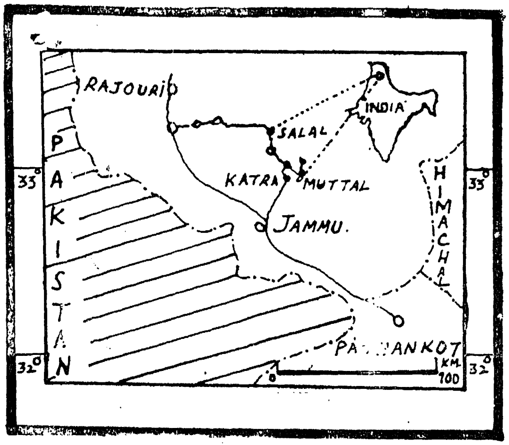
---

1. Ganju, P. N., *Proc. Ind. Acad. Sci.*, 1958, p. 345.
2. — and Pant, I. D., *Proc. Nat. Inst. Sci.*, 1963, p. 192.
3. Ramana Rao, N., *Proc. Res. Semi.*, Nagpur, 1968, p. 36.
4. — and Moiz, A. A., Paper under publication.

SOME NEW OBSERVATIONS ON THE NATURE AND ORIGIN OF CHERTS IN THE GREAT LIMESTONE FORMATION IN AN AREA BETWEEN MUTTAL AND SALAL, UDHAMPUR DISTRICT OF JAMMU, J AND K STATE, INDIA

THE present note records some of the new observations on the nature and origin of Chert bands so commonly found in the Great Limestone of Jammu (Lat. 32° 44′ N : Long. 52° E, toposheet No. 43 L/13). While working in an area between Muttal (Lat. 32° 59′ N : Long. 75° 01′ 30″ E, toposheet No. 43 p/1) and Salal (Lat. 33° 10′ N : Long. 74° 50′ E, toposheet No. $43^{k}/16$), (Map No. 1) the writer came across some well-developed bands of chert in the Great Limestone. These bands, though irregular in their pattern, exhibit a variety of brown and black colours. At places the bands are grey, buff, cream or white coloured. They are confined either in a single bed or intruded in a number of beds. Some of the chert bands are a few millimeter to several centimeters in thickness and a few meters to several meters in length.

The chert bands occur in the Great Limestone which is exposed in the form of hogback masses of mighty magnitude. Structurally these inliers are of plunging anticline, running roughly eastward, along the prevalent strike of the outer Himalaya. The limestone inliers are fitted within the Murrees and the Tertiaries. The Great Limestone is the oldest formation, whose base has not been seen in the area under investigation. On account of some economic minerals, bauxite and coal associated with some of the eocene and other rocks, the Great Limestone has attracted the attention of geologists since 1876. Among the pioneer workers, mention may however be made of Wynne[5] (1872), Medlicot[3] (1876), Middlemiss[4] (1928) and Wadia (1928, 1937) but none has dealt with the sedimentological aspects of the area in general and the nature and origin of the chert bands in particular (Figs. 1 and 2).



MAP. 1. Location Map.

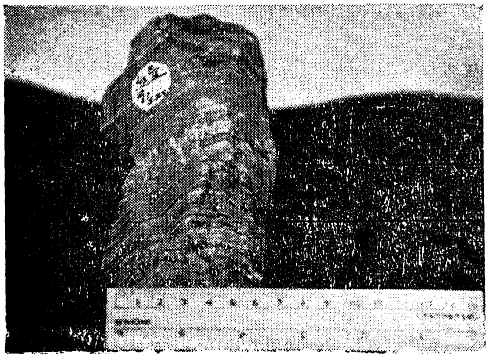


FIG. 1. Brownish and greyish chert bands in the great limestone of Riasi (Salal) area.

The detailed study of the chert bands has exhibited that the chert contains 70 to 90% of SiO_2, largely as microcrystalline to cryptocrystalline quartz with minor ratio of chalcedony. The colour and textural variations may be attributed to their respective mode of precipitation, content of carbonate and the nature of impurities. The included dolomite is anhedral and is etched and embayed by the quartz. The carbonate laminae at places are wonderfully wrapped around the bands of chert.

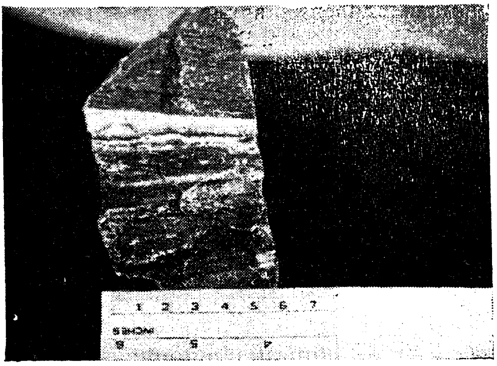


FIG. 2. White chert bands in the great limestone of Salal area.

The genetic history of the cherty matter in the limestones and dolostones offer many problems. Sharma[2] (1970) while discussing the dolostones of the area, eyplained the polygenetic origin of silica, *i.e.* Syngenetic ; penecontemporaneous, epigenetic, etc., but no specific cause has been given as to the origin of these chert bands. The present study has revealed for the first time, that the silica which precipitated into chert was derived from the pre-existisg pre-Cambrian rocks. The mobilization and distribution of the silica was mostly controlled by the pH value of the fluids, the conditions of temperature and pressure. The chert bands of the area under study are of syngenetic origin. They appear to have formed contemporaneously with the limestone or dolostones when they were redistributed by the activity of waves. It is therefore plausible to say that the fluid body that caused redistribution of silica was marine.

The material has been deposited along with the carbonates in three laterally juxtaposed environmental zones, namely the supratidal, intertidal and subtidal zones. Most of the material was deposited in the supratidal mud flat environment in the presence of hypersaline water. It is also clear that the marine water effecting silica redistribution were probably hypersaline. The silica thus carried by fresh water flocculated at the interface between fresh water and saline thereby forming the masses of silica which finally settled to form chert bands.

Abundant field evidence coupled with laboratory results have shown that the sedimentation took place in an eperic sea under warm tropical environments.

The presence of biostromes or bioberms within the Great Limestone, coated grains and oolites further support the shallow nature of the basin. It is thus concluded that the chert bands had been formed under hypersaline marine environments, where the carbonates too had been precipitated.

I am greatly indebted to Dr. T. R. Bhatia for his guidance and encouragement. Thanks are also due to Dr. T. R. Sharma, Professor and Head of the Post-Graduate Department of Geology, the Jammu University, Jammu, for his kind permission to work in the Laboratory.

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---

1. Chadha, S. K., *Curr. Sci.*, 1975, 44 (18), 665.
2. Sharma, M. R., *Unpublished, Ph.D. Thesis*, University of Jammu, 1970, pp. 118.
3. Medlicot, H. B., *Reed, G.S.I.*, 1876, 9, 53.
4. Middlemiss, C. S., *Min. Survey Report*, J and K Government, 1928.
5. Wadia, D. N., *Mem. G.S.I.*, 1928, 51, 253.
6. Wynne, A. B., *Ibid.*, 1872, 9 (2); 1.

EFFECT OF Cu-IUD ON THE LEVEL OF GLYCOGEN AND PROTEIN IN DIFFERENT ORGANS OF RAT

THE development and refinement of effective intra-uterine devices depend to a great extent on the knowledge of its operative procedure. Cu–IUD is used with a view to reducing tissue reaction, besides minimising side effects and providing better contraceptive efficacy. Significant physiological changes have been noticed in the uterus in the presence of IUCD[1-4] The metabolic changes in the uterine cornua of art with IUD have been reported[5]. But very little information is available about detrimental effect of the intra-uterine contraceptive device, on the general metabolism of the body. Tatum[6] has reported that copper content of liver, lung, etc., remains unaltered in the presence of Cu-IUD, signifying its local effect. The present observation is aimed at providing some useful information on the metabolism of glycogen and protein in various organs of the body in the presence of Cu–IUD.

25 female rats, three months old, were used in this study. Through midventral incision, bilateral insertion of copper wire was made in the uterine cornua. Another group, comprising 25 females, served as control. After fifteen days of recovery period, the rats were sacrificed and the liver, lung, pancreas, spleen, urinary bladder and the uterine cornua were taken out and placed in cold Robinson's medium. The organs were trimmed off, for fat and adhering connective tissue. The tissues from the respective organs were processed separately for biochemical estimations of glycogen and protein in the treated and controlled groups, using standard technique[7,8].

Results presented in Table I indicate that Cu–IUD fitted in the uterine cornua of rat has no significant effect on the levels of glycogen and protein in lung, kidney, pancreas, spleen and urinary bladder. However, liver, which is the main seat of storage for glycogen, shows a marked decline in the level of glycogen and protein in the treated group. On the other hand the uterine cornua treated with Cu-IUD show increased levels of both glycogen and protein.

It is evident from the results that Cu-IUD does not influence the general metabolism of the body as a whole. The protein and glycogen remain unaltered in the treated and control groups as far as lung, kidney, pancreas, spleen and urinary bladder are concerned.

TABLE I

Effect of Cu–IUD on protein and glycogen content of different organs

Organ	Glycogen µg 100 mg of tissue		Protein µg 100 mg of tissue	
	Control	Treated	Control	Treated
Liver	450·0±1·7	431·0±2·3	198·0±1·7	192·0±1·6
Pancreas	293·0±3·9	300·0±2·9	110·0±4·0	108·0±1·0
Kidney	298·0±2·4	297·0±4·0	98·0±1·2	100·0±4·5
Urinary bladder	231·0±3·8	229·0±5·4	120·0±1·9	123·0±1·4
Spleen	187·0±2·1	181·0±2·0	84·0±2·5	81·0±2·1
Lung	196·0±3·7	190·0±4·3	192·0±2·1	189·0±4·4
Uterine cornua	185·0±3·7	234·0±2·4	94·0±1·3	120·0±4·2

Cu-IUD has been reported to have better contraceptive efficacy than the other IUCDs. The antifertility effect is due to the catalysing property of copper as it participates in a number of enzymatic reactions under normal and physiological concentrations[9]. The biochemical and enzymological responses to intra-uterine devices have been noticed only in the treated cornua, the control cornua remaining as usual[10,11]. It has been suggested that the effect of IUCD is localised and is of neurogenic origin[12]. Further, it interplays through hypothalmo-hypophyseal centres. This hypothesis was upheld by Tatum[6], who reported that copper content of liver, lung, etc., remains unaltered in the presence of Cu-IUD. The effect of copper seems to be local as evidenced by normal implantation in the contralateral cornua[1].

The authors are thankful to Prof. S. M. Alam, Head of the Department of Zoology, Aligarh Muslim University, Aligarh, for providing necessary laboratory facilities.

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---

1. Zipper, J., Medel, M. and Prager, R, *Amer. J. Obstet. Gynec.*, 1969, **105**, 529.
2. Kar, A. B., *J. Sci. Ind. Res.*, 1968. **27**, 70.
3. Hugenfeldt, I., *Contraception*, 1972, **6**, 37.
4. Doyle, L. L. and Margolis, A. J., *Science*, 1963, **139**, 833.
5. Chatterjee, S. and Laumas, K. R., *Biol. Reprod.*, 1972, **7**, 307.
6. Tatum, H., *Amer. J. Obstet. Gynec.*, 1973, **117**, 307.
7. Dubois, M., Gilles, K., Hamilton, J. K., Robers, P. A. and Smith, F., *Nature*, 1951, **168**, 167.
8. Lowry, O. H., Rasebrough, N. J., Farr, A. L. and Randall, R. J., *J. Biol. Chem.*, 1951, **193**, 265.
9. Chowdhury, A. R., Chowdhury, S. R. and Kar, A. B., *Curr. Sci.*, 1975, **44**, 8.
10. Parr, E. L. and Segal, S. J., *Fertil. and Steril.*, 1966, **17**, 648.
11. Kar, A. B., Goswami, A., Kamboj, V. P., and Chowdhury, S. R., *Steroids*, 1964, **4** 159.,
12. *W.H.O. Technical Report Series No.* 397, 1968.

BETA-THALASSAEMIA TRAIT IN WEST BENGAL—A METHODOLOGICAL STUDY

THE two components of normal human haemoglobin are (1) a major one (HbA) and (2) a minor (HbA$_2$). In the detection of the beta-thalassaemia trait, rise in the HbA$_2$ proportion is usually recorded[1]. Several methods are available for its detection, involving amongst others, electrophoresis of haemoglobin on starch[2], paper[3], starch gel[4], cellulose acetate[5] and filter paper[6]. However, they are rather time-consuming and not suitable for screening large populations. Disc polyacrylamide gel electrophoresis has been found to have a much greater possibility for haemoglobin identification on a large scale[7]. A technique has been described here, based on polyacrylamide vertical disc electrophoresis with suitable concentrations gel and buffer systems, for the detection of the beta-thalassaemia trait, Since it is simple and rapid, it has been used successfully in scanning a large number of samples during population studies.

Procedure

Red cell haemolysate (10 g/100 ml) was prepared by the standard procedure. A stock solution A (acrylamide 20 g/100 ml, methylene bis-acrylamide 0·53 g/100 ml) and another solution B (pH 8·8, 17·15 g tris and 24 ml NHCl/100 ml) with 0·4 ml N, N, N′, N′-tetramethylethylene diamine, were prepared. One part of A was added to 2 parts of B, followed by one part of aqueous ammonium persulphate (0·48 g/100 ml) and the mixture was immediately transferred to glass tube (12 cm × 0·5 cm), fitted at one end with rubber caps, upto a distance of 8·0 cm. The gel was allowed to stand vertically for 30 min for setting after adding a drop of water to the upper surface to level the meniscus. A drop of bromophenol blue indicator in 2M sucrose solution was added to each tube. 20 μl of the haemolysate was applied to the gel surface after dissolving a few crystals of sucrose in the former to retard diffusion. Bridge buffer (pH 8·3, tris 0·6 g and glycine 2·38 g in 100 ml, made upto 1 litre) was poured into both the electrode chambers. Electrophoresis was carried out with a current of 4 mA per tube for a period of 120 min or upto the migration of the bromophenol blue to 6 cm from origin, at 22° C. Three sets, each containing 8 tubes, could be processed per power pack in a single day. Since HbA$_2$ moves more slowly than HbA, it can be observed as a distinct band 5 mm above the upper limit of HbA. The band given by HbA$_2$ was cut out with the help of a gel cutter, prepared by attaching two razor blades 5 mm apart in a cork. It was transferred to a tube containing 0·5 ml Drabkin's solution, macerated and kept overnight at 4° C. The HbA and other bands of haemoglobin were similarly cut out and eluted separately in 1 ml of Drabkin's solution. Measurements of the cyanohaemoglobin contents of the respective haemoglobins were made in a spectrophotometer at 420 nm (Beckmann model B 20). Originally the final dilutions of HbA and HbA$_2$ were kept equal to each other and different dilutions of HbA were tested but no significant variations were observed in the results. The readings of HbA$_2$ and the other fractions were taken in separate sittings on the same day.

TABLE I

The incidence of HbA_2 and slow moving Hb in normal poplvation in West Bengal

Sex	Total No. analysed	No. with 1–4% HbA_2	No. with 4–6% HbA_2	No. with slow moving Hb (20–40%)
Male	113	101	8	4
Female	92	79	10	3
Mean	..	2·03	4·83	24·10
S.D.	..	±0·77	±0·71	±5·21

Results

The total number of samples worked out was 205, of which 180 cases showed HbA_2 range from 1 to 4% of the total haemoglobin, as consistent with the earlier reports from India[8]. There was no difference between male and female samples. 10 females and 8 males, forming 8·78% of the total, showed a range between 4–6% of HbA_2. These were investigated fully and found to be beta-thalassaemia trait cases and form the subject of a separate communication. Seven cases, 3 females and 4 males, forming 3·36%, showed the presence of the slow moving band ranging from 20 to 40% of the total haemoglobin. Subsequent studies on the haemoglobin pattern of beta-thalassaemia and thalassaemia variants have demonstrated that the method can be used effectively in the determination of these traits.

The authors would like to express their thanks to Professor A. K. Sharma, Head of the Department of Botany, University of Calcutta and Dr. K. P. Sen Gupta, Director, Department of Pathology, Institute for Post-Graduate Medical Education and Research, Calcutta, for encourapement and facilities provided and to the CSIR (India) for the award of a Junior Fellowship to Miss M. Ajmani.

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1. Kunkell, H. C., Ceppellini, R., Muller-Eberhand, U. and Wolf, J., *J. Clin. Invest.*, 1957, 36, 1615.
2. Gerald, P. S. and Diamond, L. K., *Blood*, 1958, 13, 61.
3. Cradock-Watson, J. F., Fenton, J. E. and Lehmann, H., *J. Clin. Path.*, 1959, 12, 372.
4. Golberg, C. A. and Ross, A. C., *Clin. Chem.*, 1960, 6, 254.
5. Marengo-Rowe, A. J., *J. Clin. Path.*, 1965, 18, 790.
6. Black, M. B., Mille, H. Jr. and Wan, J., *Am. J. Clin. Path.*, 1965, 46, 483.
7. Willard, R. F., Lovell, W. J., Dreiting, B. J. and Steinberg, M. H., *Clin. Chem.*, 1973, 19, 1082.
8. Sukumaran, P. K. and Master, H. R., In: *Human Population Genetics in India*, 1974, 1, 91.

CHLOROPHYLL STABILITY OF BRINE AND FRESHWATER ALGAE

DURING our work on the effect of stress, we studied the chlorophyll stability index (CSI) of some algae. CSI indicates the stability of photosynthetic pigments when organisms are exposed to a high temperature, (65° C). In higher plants, CSI has been taken as an indicator of resistance to draught[1,2]. Purushothaman *et al.*[2] recently reported CSI of several algae, including one blue-green alga, grown in cultures.

The algae *Dunaliella salina, Schroederia setigera* (Schroeder) Lemm., *Anabaena fertilissima* and *Anabaenopsis arnoldii* were isolated from the salt lakes of Sambhar, Rajasthan. They were cultured in artificial sea water medium (Pringsheim)[3] containing 1% sodium chloride except for *D. salina* which was grown at 3% concentration. *Anabaena ambigua* and *Anabaenopsis circularis* (Watanabe strain) were cultured in Allen and Arnon's medium. *Microcystis aeruginosa* was collected from local temple tanks. CSI was determined by the method described by Purushothaman *et al.*[2]. The results are given in Table I.

The following conclusions can be drawn from our results. Among the brine algae, isolated from Sambhar salt lakes, the blue-green forms *A. fertilissima* and *Anabaenopsis arnoldii* showed high stability of their pigment at 65° C, and also the Chlorococcalean alga, *S. setigera,* to some extent. *D. salina* a Chlorophycean alga, was found to be highly sensitive to temperature. The blue-green algae generally found in freshwater habitats, *A. ambigua, A. circularis* and *M. aeruginosa* a perennial bloom of polluted temple tanks also contain pigments resistant to higher temperature.

TABLE I

Chlorophyll stability index of some algae

Algae*	Klett Units					
	In Growth Medium			In Distilled Water		
	Unheated Sample	Heated Sample	CSI	Unheated Sample	Heated Sample	CSI
1. *Anabaena fertilissima*	80	80	0	80	80	0
2. *Anabaenopsis arnoldii*	240	240	0	240	235	5
3. *Dunaliella salina*	83	48	35	83	51	32
4. *Schroederia setigera*	120	112	8	120	112	8
5. *Anabaenopsis circularis*	120	110	10	..	..	..
6. *Anabaena ambigua*	192	184	8	192	168	24
7. *Microcystis aeruginosa*	96	96	0	85	56	29

*1-4 Sambhar lake forms. 5-7 Freshwater forms.

One important observation not noted by earlier workers[2] is that the stability of the pigments in the algae is very much influenced by the medium in which they are suspended during the exposure to high temperature. When the algae *A. ambigua* and *M. aeruginosa* were suspended in distilled water and not the growth medium, the pigment stability decreased significantly (high CSI value). Suspending medium had no effect on CSI value in the case of both the green algae, *D. salina* and *S. setigera.* Determination of CSI with organisms suspended in growth media may give more reliable picture. Although, in nature, these algae may not be exposed to such a high temperature (65° C), during the mid-summer when the temperature is likely to be high, the blue-green algae form dominant flora both in Rajasthan and Banaras lakes perhaps because of their high pigment stability.

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1. Murthy, K. S. and Majumdar, S. K., *Curr. Sci.*, 1962, 31, 470.
2. Purushothaman, D., Kesavan, R., Marimuthu, T. and Oblisami, G., *Ibid.*, 1974, 43, 159.
3. Pringsheim, E. G., *Pure Cultures of Algae*, University Press, Cambridge, 1946.

PATTERN OF DORMANCY BREAKING IN SEEDS OF FEW PADDY VARIETIES

THE dormancy period for seven popular high yielding varieties of paddy, which are under cultivation in Karnataka State (India), has been assessed.

Freshly harvested seeds were subjected to the standard germination test, in the seed testing laboratory, Hebbal, Bangalore, as per the procedures laid down by the *Seed Testing Manual* (1967). Eleven tests were conducted at an interval of six days after the harvest. The test was conducted in germination room at 25–30° C.

The germination obtained in different tests are detailed below in Table I.

From the data (Table I), it is quite evident that *Kanchi* has more dormancy period than *Krishna* and *Ratna.* However, for minimum germination of 80% as per the *Minimum Seed Certification Standards* (1969) for Paddy the dormancy period is large in the varieties *IR-20* and *Kanchi* than the rest. Generally, the short duration varieties do not require a period of dormancy while the long duration varieties require a period of dormancy (Ramaiah, 1937; Narsinga Rao *et al.*, 1951). But Dore (1956) and Sahadevan (1959) found that the relationship between the duration of the crop and

TABLE I

Percentage germination obtained in different tests

Variety	Date of harvest	Days after the harvest											Days for	
		6	12	18	24	30	36	42	48	54	60	66	Complete germination	80% germination
Suma	4-11-1971	21	38	73	85	88	92	93	94	96	98	..	60	24
Krishna	9-11-1971	13	18	44	69	79	89	98	..	..	..	..	42	36
Kanchi	9-11-1971	10	15	23	27	33	43	57	65	80	83	99	66	54
Jaya	22-11-1971	4	8	39	55	61	84	90	97	98	..	..	54	36
Ratna	6-12-1971	9	22	38	46	66	91	98	..	..	..	..	42	36
Bala	6-12-1971	15	27	40	49	69	76	85	93	98	99	..	60	42
IR-20	6-12-1971	..	..	..	2	16	43	61	74	80	98	..	60	54

dormancy was not universal. In the present investigation several short duration varieties have been found to have some dormancy period.

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1. Chalam, Amir Singh, G. V. and Doughles, J. E., *Seed Testing Manual*, 1967.
2. *Minimum Seed Certification Standard*, Prepared by National Seeds Corporation, Ltd., 1969.
3. Ramaiah, K., *Rice in Madras*, Published by the Government of Madras, 1937, p. 103.
4. Narsinga Rao, M. B. V. and Shanmugasundram, A., *Madras Agric. J.*, 1953, **38**, 3.
5. Sahadevan, P. C., *Ibid.*, 1953, **40**, 133.
6. Dore, J., *Malaya Agric. J.*, 1956, **38**, 163.

SOME NEW RECORDS OF FUNGI ON MEDICINAL PLANTS

DURING the year 1973–74, the author isolated several fungi, associated with various leaf spot diseases of medicinal plants common at Bhagalpur and its suburbs. All the fungi listed in this communication are new host records from India[1-2-6]. The cultures are being identified in our Department as well as at C.M.I., Kew, England.

List of new hosts and their respective isolates

Name of the hosts	Name of the isolates
1. *Adhatoda vasica* Nees	*Alternaria alternata* (Fr.) Keissler (IMI 195059). *Chaetomium globosum* Kunze ex. Fr. (IMI 195074). *Colletotrichum capsici* (Syd.) Butler and Bisby (IMI 195078). *Botryodiplodia theobromae* Pat. *Nigrospora oryzae* (Berk and Br.) Petch *Drechslera speciferum* (Bain.) Nicot.
2. *Aloe vera* Tourn ex. Linn.	*Fusarium solani* (Mart.) Sacc. (IMI 195117) *Pestalotiopsis versicolor* (Speg.) Steyaert (IMI 195070) *Phoma sorghina* (Sacc.) Boerema, Dorenbosch and Van Kest (IMI 195069) *Phomopsis aloes-percrassae* Trinchieri (IMI 195070) *Colletotrichum gloeosporioides* Penz.
3. *Datura metel* Linn.	*Curvularia verruculosa* Tandon and Bilgrami ex. M.B. Ellis (195066 *b*) *Cochliobolus lunatus* Nelson and Hassis (195066 *a*) *Drechslera* state of *Cochliobolus specifer* Nelson (195118) *Nigrospora oryzae* (Berk & Br.) Petch.

4. *Calotropis procera* (Ait.) R.Br.	*Curvularia eragrostidis* (P. Henn.) Meyer. (IM 195063) *Chaetomium globosum* Kunze ex. Fr. (IMI 195074) *Fusarium equiseti* (Corda) Sacc. (IMI 195068) *Acremonium rutilum* Gams. (IMI 195083) *Nigrospora oryzae* (Berk & Br.) Petch.
5. *Vitex negando* Linn.	*Colletotrichum* state of *Glomerella cingulata* (Stonem) Spauld and Schrenk. (IMI 195077) *Drechslera* sp. (IMI 195063) *Curvularia tritici* Kumar and Nema (IMI 195082) *Alternaria alternata* (Fr.) Keissler (IMI 195063) *Curvularia lunata* (Wakker) Boedijn.

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---

1. Butler, E. J. and Bisby, G. R., *The Fungi of India,* Issued by I.C.A.R., New Delhi, 1931–52.
2. Pathak, N. C. and Ghosh, R. N., *Fungi of Uttar Pradesh, Bull. Nat. Bot. Garden,* Lucknow, India, No. 62, 1962.
3. Sarbhoy, A. K., Lal, G. and Varshney, J. L., *Fungi of India,* Published by Navyug Traders, New Delhi, 1967–71, p. 150.
4. Tandon, R. N. and Chandra, S., *Supplement to the List of Indian Fungi,* Botany Department, Allahabad University, 1957–62.
5. Tailak, S. T. and Rao, R., *Fungi of India,* Supplement 2, 1962–67.
6. Vasudeva, R. S., *Ibid.,* Supplement 1, Issued by I.C.A.R., New Delhi, 1953–57.

SHORT SCIENTIFIC NOTES

Occurrence of Collar Rot of Fenugreek in Karnataka

Fenugreek (*Trigonella foenum-graecum* L.), a common leafy vegetable, was found to die prematurely in an experimental plot, in December 1975, at the Agricultural College, Hebbal. The affected plants were characterized by a sudden wilting of the seedlings, distinctly seen in patches in an otherwise healthy plot. The symptoms at the collar region was girdling of the epidermal tissue associated with brown discolouration. The roots were shortened and dark.

Isolations made from the affected parts yielded consistently a non-sporulating, septate fungus with abundant brown sclerotia, loosely formed among and connected by mycelial thread. The fungus was brought into pure culture and was then grown on corn meal sand agar. A week old of this culture was mixed with sterile soil so as to give different inoculum levels starting from 1% upto 10% of the soil (w/w). Surface sterilized seeds of Fenugreek were then sown, 100 each per pot. Ten days later observations were recorded. In the pots mixed with inoculum, the sprouted seeds were affected by the fungus which resulted in complete death, as against 90% germination in uninoculated check. There was cent per cent death at all the inoculum levels tried. The same fungus was reisolated from the affected sprouts.

The pathogen was identified as *Rhizoctonia solani* Kuhn. The culture has been deposited in the culture collections of the Department of Plant Pathology, University of Agricultural Sciences, Bangalore. bearing accession No. 142. Hitherto there is no record of *R. solani* on Fenugreek from India and this constitutes first report.

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Veronaea simplex from India

The hyphomycete *Veronaea simplex* Papendorf was described[1] in 1969 from leaf-littler and top soil of a mixed *Acacia karroo* community Potchefstroom, Transvaal, S. Africa. A fungus which matches the description of *V. simplex* perfectly has been isolated by the author twice, first in March 1962 and then in August 1975 from soil of a sal (*Shorea robusta*) forest of Gorakhpur, U.P. Both isolations were made in soil-plates prepared with Martin's peptone-dextrose agar medium containing streptomycin and rose bengal[2]. The species is reported here for the first time from India. A dried culture has been deposited in the Commonwealth Mycological Institute, Kew, as IMI 197680.

The author wishes to thank Dr. M. B. Ellis of C.M.I., Kew, for confirming the identity of the fungus, and Prof. K. S. Bhargava for laboratory facilities.

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1. Papendorf. M. C., *Trans. Brit. mycol. Soc.*, 1969, 52, 483.
2. Martin, J. P., *Soil Sci.*, 1950, 69, 215.

Leaf Spot of Cacao

'Black pod' disease caused by *Phytophthora palmivora* Butl.[1] and 'charcoal pod' rot caused by *Botryodiplodia theobromae* Pat. and *Macrophoma* sp.[2], in cacao (*Theobroma cacao* L.) have been reported so far in India. Incidence of a leaf spot disease was first noticed in 1972 in plants of 'Forestero' and 'Criollo' varieties of cacao grown in CPCRI farms at Kasaragod (Kerala) and Vittal (Karnataka), and also in some private plantations near Sullia (Karnataka).

The disease appears as minute yellowish specks which are later discernible as circular reddish brown lesions often with a chlorotic halo. A mature lesion may reach a size of 2–4 mm and occasionally two or more adjacent lesions may coalesce forming irregular necrotic patches. In severe cases crinkling of the leaf also is noticed. The mature lesions being dry and brittle, at times, may either drop off partially or be blown away completely by wind resulting in 'shot-hole' formations. The fructifications are noticed mostly on the lower side of the lesions as greyish white masses. The disease is seen throughout the year but the intensity is more during October–January when a temperature range of 19–33° C and R.H. range of 77–98% prevail. The disease was not found to do much damage to the leaf. However, in serious cases much of the lamina will be involved resulting in impairment of the functional photosynthesis. *Colletotrichum*, sp. was isolated from the affected leaves. The fungus was also isolated from the pods affected by rot. The isolates were found to be cross inoculable giving positive pathogenicity on leaves and pods.

When artificially inoculated the symptoms appear in 4–6 days after inoculation. The isolate sporulates moderately on potato-sucrose-agar and abundantly on oats agar media. The fungus was identified as *Colletotrichum gloeosporioides* Penz. and the same has been confirmed by CMI London (IMI. 184312). So far two species of *Colletotrichum* have been recorded on cacao : *C. crassipes* (Speg.) V. Arx causing cacao wilt[3] and *C. gloeosporioides* Penz. causing pod rot, leaf blight and die-back[4,5] in Brazil. We record the occurrence of leaf spot of cacao caused by *C. gloeosporioides is* India for the first time.

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1. Ramakrishnan, K. and Thankappan, M., *South Ind. Hort.*, 1965, 13, 33.
2. Nambiar, K. K. N. and Radhakrishnan Nair, R., *Indian Phytopath.*, 1972, 25, 595.
3. Ram, A. *et al*, *Rev. Theobroma*, 1973, 3, 33.
4. Desrosiers, R. and Diaz. J. M., In *Conferencia Interamerica de cacau* ; 6 .a.; Salvador, Bahia, Brasil. 1956. Bahia, Brasil Institute de cacau, 1957, 331.
5. Diaz, F., *Phytopathology*, 1967, 57, 7.

Pteris excelsa Gaud.—A New Fern Record for Jammu and Kashmir State

Perusal of literature has revealed that *Petris excelsa* Gaud. which is common in Kumaon Garhwal, Dalhousie, Sikkim and Naini Tal has not been reported from the Jammu and Kashmir State so far. The present report based upon the specimens collected from Poonch (Bufliaz Forest, 2000 m) along slow moving waters ; and from Bhaderwah (Nandoun Nallah, 1750 m) in J and K State, under the collection Nos. 1469 and 1116 of Harvender thus constitute a new fern record for the State. The pressed specimens have been deposited in the Botany Department of Government Degree College, Poonch.

Brief Description :

Petris excelsa Gaud. in *Freyc. Voy. Bot.* 388. 1829.

Fronds 1–1·20 m lang, submembranaceous, light green to pale in colour, pinnate; pinnae large, remote, numerous, lanceolate, sessile, 30–45 cm long, lower one's long petioled, all deeply cut nearly to the costa; segments 4–10 cm long, linear-lanceolate, obtuse, serrated, subfalcate; Veins free, forked near the middle; sori continuous from the base, almost to the apex, broad; stipes very stout and flexuous, rachises bright castaneous.

Grateful thanks are due to Prof. S. S. Bir, Head, Department of Botany, Punjabi University, Patiala, for identifying the specimens and to Dr. A. K. Koul, Department of Bio-Sciences, Jammu University, for going through the manuscript.

Department of Bio-Sciences, HARVENDER KIRN.*
Jammu University, JAGDISH SHARMA.**
Jammu 180 001, *May* 31, 1976.

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Natural Occurrence of Potato on Virus-Y *Datura metel*

A disease of *Datura metel* Linn. characterized by severe mosaic, malformation and distortion of leaves and in some cases crinkling of leaf lamina, resulting in filiformed leaves was found in the fallow lands in Bangalore. Flowers also showed distortion and the fruits were partially or wholly devoid of spines.

The disease was readily transmissible by sap to *Datura metel* Linn., *Capsicum annuum* Linn. var. California wonder, *C. frutescens* Linn. var. Tabasco, *Nicotiana tabacum* Linn. var. White Burley, *N. glutinosa* Linn., *Solanum tuberosum* Linn., *S. nigrum* Linn. and *Petunia hybrida* Vilm. producing systemic infection. The virus produced chlorotic local lesions on inoculated leaves of *Chenopodium amaranticolar* Coste and Reyn. and necrotic local lesions on 'A_6' hybrid potato. The aphids *Myzus persicae* Sulz. and *Aphis gossypii* Glover could transmit the virus from infected to healthy *D. metel* and also to *C. annuum* var. California Wonder. The virus was not seed borne.

The virus had a thermal inactivation point of 55–60° C, dilution-end-point of 1 : 500 to 1 : 1000 and longevity *in vitro* for 4 days at room temperature. It was purified by polyethylene glycol method[5]. The purified preparation was highly infectious and the electron microscopic studies revealed the presence of flexuous rods with 733 mμ length and 13 mμ breadth.

There are several reports of virus diseases reported on different species of *Datura* from India[1,2,4,6,8,9]. The virus under study differs from all these viruses in its host range, physical properties and also being able to produce local lesions on *Chenopodium amaranticolar* and A_6 hybrid potato. Feldman and Gracia (1972) for the first time reported that *D. feroxa* Linn., as a natural host of PVY in Mendoza, Argentina. The present observations made on the virus disease of *D. metel* shows that it has the host range, physical properties and insect vectors similar to that of PVY. Further, electron microscopic studies also indicated that the virus is a flexuous rod measuring 733 mμ length and 13 mμ breadth which falls in the range of PVY particles[7]. From the above studies the present virus could be considered as PVY naturally occurring on *D. metel* not reported earlier.

Grateful thanks are due to Dr. H. C. Govindu, for his keen interest and encouragement throughout the course of this investigation and to Dr Rama Mohan for taking Electron micrographs.

Department of Plant R. D. V. J. PRASAD RAO.
Pathology, R. C. YARAGUNTAIAH.
University of Agricultural Sciences,
Hebbal,
Bangalore 560 024, *February* 24, 1976.

1. Capoor, S. P. and Verma, P. M., *Curr. Sci.* 1948, 19, 151.
2. — and —, *Indian J. Agric. Sci.*, 1952, 22, 303.
3. Feldman, J. M. and Olga Gracia, *Phytopath. Z.*, 1972, 73 (3), 251.
4. Garga, R. P., *Indian Phytopathology*, 1958, 11, 133.
5. Hebert, T. T., *Phytopathology*, 1963, 53, 36.
6. Padmanabhan, C. and Kandaswamy, T. K., *Madras Agril. J.*, 1973, 60 (7), 548.
7. Varma, A., Gibbs, A. J., Woods, R. D. and Finch, J. T., *J. Gen Virol.*, 1968, 2, 107.
8. Verma, G. S. and Verma, H. N., *Indian Phytopath.*, 1963, 16, 368.
9. Yaraguntaiah, R. C. and Govindu, H. C., *Mysore J. Agric. Sci.*, 1972, 6, 352.

Helminthosporium Leaf Spot on Cotton

GIZA–7, the cotton (*Gossypium barbadense* L.) grown in Guntur and Prakasam Districts was severely affected by a leaf spot disease, at the age of 4 to 5 months during January, 1975. Numerous, circular, light red to dark purple spots with ashy centres, measuring 1 to 10 mm in diameter were observed on the leaves.

Examination of the infected leaves showed constant association of *Helminthosporium* sp. in the diseased tissues. The fungus was brought into culture on P.D.A. but no sporulation was observed even after a month. Artificial inoculation of the GIZA–7 plants of 4 months old, with 12 days old culture of the fungus, produced typical symptoms of the disease in 6 days. Examination of the spots revealed the presence of *Helminthosporium* sp. and the same was brought into pure culture. A comparison of the morphological characters of the present fungus with those of *Helminthosporium gossypii* reported from Porto Rico by Tucker[1] (1926) showed close resemblances except in the length of conidia. Hence the present fungus is tentatively identified as *Helminthosporium gossypii* Tucker.

The authors are grateful to Dr. S. N. Rao, Principal, Agricultural College, Bapatla, for his keen interest and for providing necessary facilities.

A.P. Agricultural University, S. RAMAPANDU.
Dept. of Plant Pathology, K. ABBAIAH.
Agricultural College, SHAIK MOHIDDIN.
Bapatla, *April* 5, 1976.

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1. Tucker, C. M., *J. Agric. Res.*, 1926, 32, 391.

Leaf Blight of Green Gram Caused by *Curvularia eragrostidis*

A leaf blight of green gram (*Phaseolus aureus* Roxb.), affecting the tips, was observed in the later part of October, 1975, in the Research Farm of Assam Agricultural University, Jorhat. Initially the disease manifested as small, irregular brown spots at the apices usually about 1–4 mm long which increased in size, became necrotic and covered with copious growth of fructifications.

Isolations from the affected regions consistently yielded a *Curvularia* sp. Pathogenicity was established by inoculating both wounded and unwounded leaves by spraying spore suspension of the fungus from a one-week old culture: symptoms developed after 3–4 days on wounded and 8–10 days on unwounded leaves.

In a field survey the disease was found to attack all the 11 cultivars grown in a varietal trial but with different intensities as shown within the paraentheses (% of plants infected)—Madira (80), ML 24 (75), H-70-16 (72), Sirsa (70), ML 4 (64), T44 (61), PS 11 (55), K 85 (50), PS 2/1 (44), S 9 (58), K 851 (30).

The fungus has been identified as *Curvularia eragrostidis* (P. Henn.) J. A. Meyer. Among the leguminous plants, only the members of *Arachis* have been categorized as hosts of this fungus[1] and therefore, *P. aureus* is a new addition. This pathogen has recently been recorded on pineapple from Assam by Roy[2].

The specimen has been deposited to the H.C.I.O., New Delhi, under the accession No. 31970.

Thanks are due to Shri L. Upadhaya, Pulse Geneticist, for referring the problem to us.

Mycology Research Section, B. BARMAN.
Assam Agricultural University, A. HYDER.
Jorhat 785 013, *November* 24, 1975. A. K. ROY.

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1. Ellis, M. B., *Dematiaceous Hyphomycetes*, C.M.I., Kew, 1971, pp. 608.
2. Roy, A. K., *Curr. Sci.*, 1975, 44, 467.

A New Host Record of *Pringsheimia* sp. on Sannhemp (*Crotalaria juncea* L.)

During the month of September, 1965, some brown spots bounded by veins and veinlets were observed on the lamina of mature green leaves of *Crotalaria juncea*. The spots were found to be associated with a fungus, hitherto not reported on this crop.

Microscopic observation showed that these were globose, black, erumpent perithecia scattered all over the necrotic spots. The outer perithecial wall was thin and pseudoparenchymatous. Each perithecium contained a number of pyriform (or slightly clavate), hyaline asci with a thin but tough wall. Each ascus was with 8 spores, which were hyaline, somewhat thick walled, 4–7 celled, muriform with longitudinal septum appearing in one or two cells only.

The perithecia *in vivo* were 128·0 μ to 137·0 μ in diameter. The asci measured 41·3 μ to 48·0 μ long and 24·1 μ to 27·5 μ broad. Ascospores measured 24·1 μ to 27·5 μ long and 8·2 μ to 10·3 μ broad, 4–7 celled muriform.

The fungus was easily isolated on standard potato dextrose agar. Initially the developing mycelium forms a brown appressed mat which gradually turned into mouse-grey by the fourth day and were mature after seven days of growth.

The perithecia *in vitro* measured 146·0 μ to 154·8 μ in diameter. The asci measured 48·16 μ to 89·44 μ long and 24·0 μ to 36·1 μ broad. The ascospores measured 26·4 μ to 29·2 μ long and 9·5 μ to 11·0 μ broad, 4–7 celled muriform with somewhat thick wall.

The only report of *Pringsheimia* from India, which the author is aware of, published by Kapoor and Gill (1961).

The author is grateful to Dr. S. P. Ray Chaudhary, former Head of Division of Mycology and Plant Pathology, I.A.R.I., for identifying the pathogen.

Jute Agricultural C. B. P. MISHRA.
Research Institute,
Barrackpore, *October* 28, 1975.

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1. Kapoor, J. N. and Gill H. S., "Notes on Indian Ascomycetes—I," *Indian Phytopathology*, 1961, 14, 149.

REVIEWS AND NOTICES OF BOOKS

Structure of the Nucleus. By M. A. Preston and R. K. Bhaduri. (Addison-Wesley Publishing Company, Advanced Book Program, Reading, Massachusetts 01867), 1975. Pp. xiv + 693. Price $ 29.50.

This book is a revision of M. A. Preston's 1962 book—*Physics of the Nucleus.* It is intended to be useful as a reference volume for research physicists. Graduate students engaged in a first detailed examination of nuclear physics should also find it useful. In the preface to the earlier book, it was said that nuclear physics had advanced almost far enough that one could attempt to write a book on a deductive basis, stating first the fundamental laws determining the interaction of nucleons and then deducing the properties of the nucleus. If this view was realistic in 1960, it should be even more capable of fulfilment in 1975. But we are now only in a position to appreciate the difficulties encountered in completing some of the links in the chain of deductive reasoning. Though we know a great deal more about the two-nucleon force, it is still not completely understood. Our present-day knowledge of the nature and role of many-body forces and the structure of the nucleons themselves has many gaps. Many aspects of the behaviour of nuclei are best discussed in terms of models whose limits of validity are not fully understood in terms of a more fundamental theory. The authors of the present book have tried to emphasize the fundamental approach which unify the concepts used in describing nuclei. But this book, like its predecessor, has remained semideductive.

The book is divided into three parts : (1) Fundamental properties of nuclei, (2) Nuclear models and (3) Alpha-disintegration and Fission of Nuclei. The earlier book had five parts : The parts entitled E.M. properties of nuclei and nuclear reactions have been dropped from the present volume. The part entitled "Particle Radioactivity" in the earlier book becomes "Alpha-disintegration and Fission of nuclei" in this book. The chapter on beta-activity is dropped from the present book. As in the earlier book the authors have given references and problems at the end of each chapter.

The understanding of the subject-matter dealt with in this book requires a knowledge of basic quantum mechanics. Though authors assume that the reader has had a prior introduction to nuclear phenomenology, the disccussion of each topic proceeds *ab initio* and makes no essential use of unstated facts of nuclear physics. The discussion throughout the book is more theoretical than experimental.

The reviewer feels that some of the topics like beta-decay, inner bremsstrahlung, heavy ion interactions, pion-nucleon interaction and nucleon-nucleus scattering should have been included in the book and more detailed discussion of the muonic atoms would have been useful. Nevertheless the book is welcome, as a reference book, to research physicists and post-graduate students. The general get-up of the book is quite good.

B. Sanjeevaiah.

Guidebook to Stereochemistry. By F. D. Gunstone. (Longman, London), 1975. Pp. xii + 112. Price £ 2.95 p in U.K.

The concept that molecules exist in a three-dimensional form, put forward independently by Le Bel and by van't Hoff nearly a century ago, is the cornerstone of stereochemistry. In this little "guidebook" under review, Prof. Gunstone has introduced the basic concepts of this subject in a way that should appear to both the beginner as well as others who are more familiar with the subject. There are chapters on *cis-trans* isomerism, enantiomerism, conformation and dynamic stereochemistry. The author points out at the beginning that a study of this book should enable the reader to use correctly the terms : chiral, achiral, and prochiral ; chiral centre, chiral axis, and chiral plane ; chair, boat, and twist conformation; *cis-trans*-isomerism; enantiomer, diastereoisomer, meso compound, and racemic form ; R, S, E, and Z ; sawhorse, Newman and Fischer projections ; conformation, eclipsed, staggered, synperiplanar, antiperiplanar, synclinal, and anticlinal. The author has certainly succeeded in his objectives. The structure of cyclododecane on p. 69 is not correct. Also the representation of all axial bonds by solid lines and all equatorial bonds by broken lines may confuse the reader as one is accustomed to associate solid lines with β-substituents and broken lines with α-substituents. However, these are minor drawbacks and the book will be useful to all those who wish to have a basic knowledge of modern stereochemistry.

K. M. Sivanandaiah.

Agricultural Systems. *Vol. I, No. 1 : An International Journal.* Edited by C. R. W. Spedding. (Applied Science Publishers, Rupple Road, Barking Road, Essex, England), 1976. Pp. 83. Annual Subscription £ 15/– inclusive of postage (4 issues per volume).

The publication under review is the first quarterly issue of the new Journal entitled *Agricultural Systems* brought out in Great Britain. The object of the Journal as the Editor states is to provide a focal point for thinking about agricultural systems which is a bridge-building subject to be studied with an overall purpose of helping to define and solve problems of practical importance in agriculture.

Agriculture is said to be the mosaic of arts and sciences embodying such diverse subjects as biology, management, economics and social sciences. The interactions among these disciplines make the practice of agriculture a highly complex proposition though basically it is a practical business world over, involving not only livelihood of sizeable population but also feeding of all human beings and their livestock.

It is rightly said, if applied agricultural research has to have practical relevance, the understanding and definitions of problems and systems need considerable refinement. There is a necessity for clarity in respect of statement of problems, because the answers, when they go back, have to fit into a complex system, full of interactions with other components.

In his paper on Economics of Systems Research, Dillion states that systems approach provides a better view of agriculture system and also facilitates purposeful research management in terms of good achievement and choice between research alternatives. Discussing information system, for individual farm as exemplified by a pig herd, Blackie shows that a term approach to problem solving, offers a significant pay-off as opposed to the traditional random piecemeal approach. Smith and Williams in their study on the effects of deferred grazing of annual Medeterranian pasture on sheep production, suggest the use of system analysis approach in order to obviate the uncertainty associated with the expected returns. Shedding and Brockington lay emphasis on defining the role of experimentation to the object of the experiment. Baker and Horrocks suggest a mathematical model that simulates energy and gas exchange at the plantair interface, to help analysing corn production.

The integrated approach in problem-solving in India is an inescapable necessity, if we have to achieve production targets to meet the growing needs of the country. The development of the concept of agriculture systems in problem-solving will be of great interest to all concerned with agriculture. This concept is yet to be adopted in the country.

H. R. ARAKERI.

Annual Review of Phytopathology (Vol. 13). Edited by K. F. Baker, G. A. Zentmyer and E. B. Cowling. (Annual Reviews, Inc., Palo Alto, California, USA), 1975. Pp. 1–412. 22 × 14·5 cm. Price : USA. $ 15.00. Elsewhere : $ 15.50.

With the rapid strides in the various branches, reviews on particular disciplines of Botany have played a significant role for a better understanding and from these points of view, the *Annual Review of Phytopathology* has subserved a very useful function. Already, twelve volumes have been published and practically all the papers are strongly international in content written by several authors from 23 countries.

The first article on Fungi and Fungicides, the story of a non-conformist, is very ably written by James G. Horsefall of the Connecticut Agricultural Experimental Station, New Haven and a beautiful photograph of his oil painting is provided as a frontispiece. This is followed by a series of extremely well-written articles like Historical perspective, Appraisal of plant disease, Pathogens, Morphology and anatomy, Physiology of host-pathogen interaction, Epidemiology, Influence of environment, Chemical control, Biological and cultural control, Breeding for resistance and these special topics are written by specialists in the field. While most of the contributions are written by single authors, two or more authors have contributed on the articles on Morphology and anatomy, Physiology of host-pathogen interaction, Influence of environment and Pathology ; seven authors have written very authoritative and useful reviews on Pathogens and it appears to be quite right that great importance is given to this aspect covering nearly 77 pages. Another redeeming feature of this review is the last chapter on Indexes which cover in a detailed manner an author and subject index, cumulative index of contributing authors of vols. 9–13 and a cumulative index of chapter titles for these volumes. The coverage of literature of each article is complete and every article reviews all the recent aspects of Phytopathology in an able manner. The extremely well-thought-of illustrations, tables and photographs are very suggestive.

On the whole, this *Annual Review of Phytopathology* is an excellent contribution and as usual it has maintained the high standards of the previous volumes. Without any hesitation, it must find a place in all departments and institutions where studies on Phytopathology are carried out.

K. Subramanyam.

Enzyme Structure and Function. By S. Blackburn. (Publishers: Marcel Dekker, Inc., New York and Basel), 1976. Pp. viii + 528. Price: $ 39.50.

This volume constitutes the fourth in the series; the publishers are bringing out a series of Text-books and Monographs on Enzymology. Two features of special nature are noted: The matter is printed by photo offset and there is not a single reference of the author himself. It is interesting to know that the author's "current work centers on scientific writing and documentation". Thus, a overview of this vast and fast developing field has been presented in these 500 and odd pages by a person who put all the facts together in a cogent fashion instead of an overbearing personal bias if the author himself had directly contributed to the field. The author has chosen all hydrolytic enzymes on which extensive work has been carried out. These are: chymotrypsin, trypsin, elastase, carboxypeptidases, subtilism, papain, staphylacoccal proteinase and nuclease, ribonuclease and lysozyme. These are excellent records of the knownledge gained on these enzymes with respect to specificity, active site, structure, reaction mechanism, binding of substrate, and 3-dimensional structure. These are padded in the beginning by the experimental techniques and in the end, by current status on the subject with respect to structure, mechanism and evolution of enzymes. No doubt these are excellent presentations and those interested in this area will find this book an extremely useful reference work. Those wishing to know the trends of research on these enzymes are well advised to read this book instead of multiple reviews and original papers. Reading through the pages and following the general thrust in the studies on mechanism of enzyme action, the reviewer had noted a large emphasis on the active site and a corresponding neglect of the large chunk of the rest of the protein. In fact, the author concluded on the item of what may be called equivalent synthetic active site of myoglobin, purported to mimic fully the natural process and considered this as a foreshadow of the exciting possibilities ahead. Finally, in view of the restrictive nature of the type of enzymes covered, the tittle of "Structure and Function of Hydrolytic Enzymes" would have been more appropriate.

T. Ramasarma.

Mechanics of Machines. By G. H. Ryder and M. D. Bennet. (The Macmillan Press Ltd., London), 1975. Pp. vii + 227. Price: £ 8.95.

This is an introductory book in the mechanics of machines for students in polytechnics and those working for higher national certificate and higher national diploma courses in U.K. As a book written at the polytechnic level, it is an excellent synthesis of the traditional subjects of mechanics including dynamics and statics, kinematics, including mechanisms the marginal subject of balancing of machines. Topics also include vibrations of single and multi-degree freedom system, lateral vibration including whirling speeds and elements of control systems. The authors have made a judicious selection of topics in the above area to produce a book acceptable at the polytechnic level. Throughout the book, theory is illustrated by means of engineering applications over a wide field. Numerical examples have been carefully selected to give a realistic appreciation of the actual system parameters and performance. The book is in S.I. units throughout.

Although the authors claim that the book forms a solid base for more advanced studies for the 1st and 2nd year students of the degree course the reviewer is positive that it does not adequately cover the material for those majoring in mechanical engineering speciality. In the reviewer's opinion it is an excellent introduction in theory of machines for students majoring in civil, electrical, electronic, chemical engineering except mechanical engineering.

As regards the suitability of the book for polytechnics in India, the reviewer feels the book is a little highbrow because the fundamental background of the students in polytechnics in India may not be adequate to grasp the concepts.

One thing that is definitely against the book is its prohibitive cost. The book is priced at £ 8·95 (hard cover) and £ 4·95 (paper back). The get-up of the book set in 10/11 pt IBM is excellent.

P. Srinivasan.

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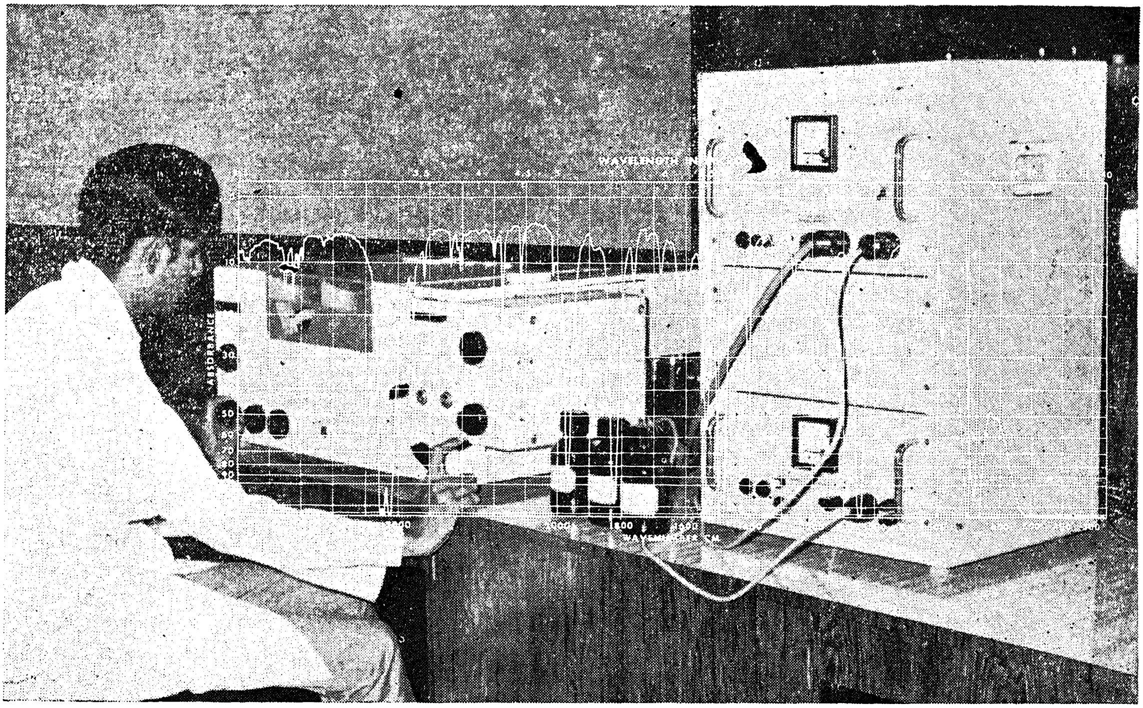

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BETA DECAY OF ^{166}Ho

K. VENKATA RAMANIAH, V. LAKSHMINARAYANA AND K. VENKATA REDDY
Laboratories for Nuclear Research, Andhra University, Waltair

ABSTRACT

The 27 hour beta decay of ^{166}Ho has been studied with a 35 cc Ge (Li) detector and an intermediate image beta ray spectrometer for an accurate analysis of the gamma spectra and beta spectra to estimate precise relative intensities of gamma lines and to search for any new gamma lines. The β spectrum is used to determine the exact end-point energies, intensities and log ft values of different beta groups. The possibility of a beta feeding to the 2^+ excited state at 787 KeV of ^{166}Er is ruled out.

INTRODUCTION

THE ground state decay of ^{166}Ho (27 *h*) populates the ground state and five excited states of the even-even deformed nucleus ^{166}Er. The gamma emission in the decay of ^{166}Ho was investigated by Marklund *et al.*[1] and Cork *et al.*[2] Cork *et al.* reported a 470 KeV gamma ray observed in their scintillation spectrum. They did not present any spectrum and gave no value for the intensity of the 970 KeV gamma ray A later investigation was by Burson *et al.*[3] using a 2 c.c. planar Ge (Li) detector, the results of which, in general, did vary from the earlier reports. There are also considerable discrepancies in the reported results on the end-point energies, intensities and log ft. values of different beta groups of ^{166}Ho.

The availability of a large volume Ge (Li) detector in this laboratory prompted the present investigation on the relative gamma intensities of gamma rays, in the decay of ^{166}Ho. The gamma spectrum in this decay is reinvestigated with a 35 c.c. Ge(Li) detector with an aim of performing more accurate measurements on the relative gamma intensities and to search for the possible existence of weak gamma lines hitherto unobserved and to determine the extact beta group emergies intensities and log ft values with the help of the beta spectral data from the meqsurements on an intermediate image beta ray spectrometer.

γ-Spectrum: *Experimental details and results.*—The stable isotope of helmium which has a thermal neutron cross-section of ∼65 barns was irradiated at the Research Reactor, CIRUS of Bhabha Atomic Research Centre, Bombay. The isotope was obtained in the form of helmium trichloride in hydrochloric acid solution. The gamma spectra are recorded with a ND-512 analyser at regular intervals of 27 hours to identify the gamma rays belonging to the decay of ^{166}Ho. Ten days after the present work, a background run is taken over the entire energy range to obtain the background as well as the presence of long lived impurities in the source. The detector was calibrated earlier[4] for relative photopeak efficiency using gamma rays from ^{152}Eu and ^{124}Sb. These data are used to deduce the relative photon intensities. The absolute intensity of the 1367 KeV transition is assumed from the earlier accurate measurement of Gline *et al.*[5]. The K-conversion coefficient and K/L ratio of the 80 KeV transition is taken from the Nuclear data Sheets and used in the calculation of the transition intensity of the 80 KeV gamma ray.

Figure 1 shows a typical gamma spectrum recorded in one of the runs. As can be seen from the gamma spectrum, no new gamma rays are observed following the decay of ^{166}Ho. A careful search is made for detecting the presence of possible weak gamma rays corresponding to the decay of 1830 KeV state to the 1460 and 1663 KeV states, but no such gamma rays have been observed in the present work.

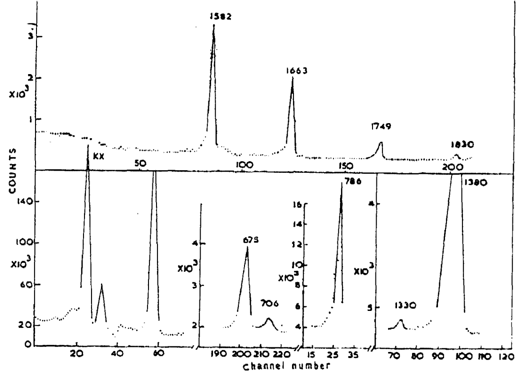

FIG. 1. Gamma spectrum of ^{166}Ho taken for different regions with 35 co Ge (Li) detector.

Table I summarises the present results on the relative gamma intensities along with the earlier results; there is a good agreement between the two sets of data except in the case of the intensities of the 1830 and 1663 KeV gamma transitions. The present results failed to locate a beta branch of observable intensity to the 2^+ excited state at 787 KeV which is also confirmed from the present beta-gamma coincidence measurements.

Beta Spectrum: *Experiment and Results.*—A Siegbahn-Slatis beta ray spectrometer with plastic well type detector used in the investigation is described

TABLE I

Relative intensities of gamma rays in the beta decay of ^{166}HO

Gamma energy (KeV)	Relative photon intensities	
	Present work	Burson *et al.*[3]
80·6	6·55 ±0·3	6·2 ±0·4
675·0	0·032 ±0·002	0·03 ±0·002
707·0	0·021±0·001	0·019±0·003
786·0	0·011 ±0·005	0·015 ±0·003
1380·0	0·93 (std.)	0·93 (std.)
1582·0	0·20 ±0·01	0·19 ±0·009
1663·0	0·0092±0·004	0·12 ±0·006
1749·0	0·028±0·0017	0·031±0·001
1830·0	0·0075±0·005	0·0093±0·0007

elsewhere[6] and is suitably modified[7] for $\beta-\gamma$ coincidence measurements. The sources used are of 200 μg/cm^2 thick and 2 mm in diameter and are prepared by evaporatiion of the source on zapon films. The beta spectrum is scanned roughly in steps of 12 KeV upto 1850 KeV. The background is taken at every measurement point. In the present measurement the first inner beta group in coincidence with the 80 KeV gamma ray is scanned. The Fermi-Kurie analyses of the coincidence and the cross-spectra has resulted in the end-point energies of 420 ± 4, 1771 ± 2 KeV and 1845 ± 2 KeV for three beta groups. A further subtraction of the 420 KeV beta group from resulting gross-spectrum has revealed a fourth beta group (which is not shown in Fig. 2) with an end-point energy of 230 ± 4 KeV. No evidence could be found for a weak beta with an end-point energy of 1070 KeV feeding the 787 KeV level which has also been confirmed from our gamma spectrum. The Fermi-Kurie analysis of the ^{166}Ho beta spectrum is shown in Fig. 2.

Half life of ^{166}Ho.—The current setting of the present spectrometer is adjusted at 1600 KeV and the count-rate is followed for four half lives. A least square fit of the data yielded a half life of (27 ± 0·04) hours. This also ensures the absence of any other foreign activities in the sample.

Beta intensities and log ft values.—The intensities of the beta branchings are deduced from the present relative gamma intensities assuming a 0·93% decay of ^{166}Ho (0$^-$) ground state to the 0$^+$ excited state at 1460 KeV. The log ft values of the beta transitions are calculated using the recent tabulations[8] of log ft values, employing the beta end-point energies and the present gamma intensities. The results are presented in Table II along with the earlier results. The log ft values of the beta transitions leading to the two 0$^+$ states of ^{166}Er are quite different. Further the log ft. value of the beta branch to the 1830 KeV state is indicative of an allowed sature, thus establishing the 1$^-$ assignment to the 1830 KeV state. The present results are incorporated in a decay scheme shown in Fig. 3.

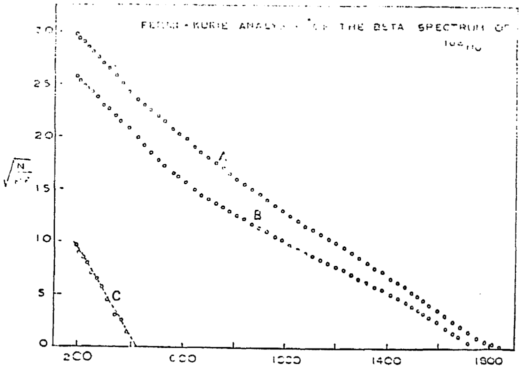

FIG. 2. Fermi-kurie plots of the three beta groups of ^{166}Ho uncorrected for shapes A, F.K plot of the total spectrum. B, F.K. plot of the coincidence spectrum of (0$^-$ → 2$^+$) transition. C, F.K. plot of the resulting after the subtraction of the intense two beta components.

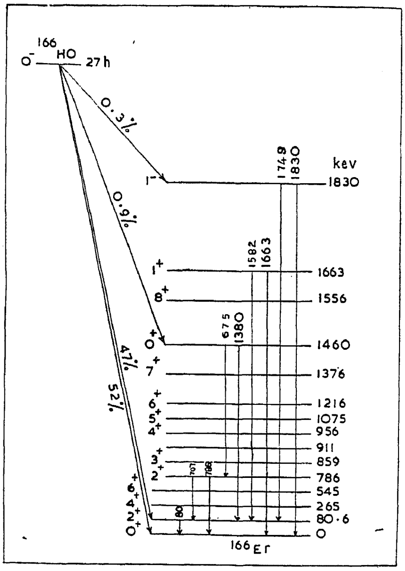

FIG. 3. Decay scheme of the levels of ^{166}Er populated from ^{166}Ho (27·0 h) incorporating the present results.

TABLE II

Data on the Beta groups in the decay of ^{166}*Ho*

Present work			Sunyar[9]		Graham *et al.*[10]		Cork *et al.*[2]		Funke *et al.*[11]	
Energy (KeV)	Int. %	Log ft.	Energy (KeV)	Int. %	Energy (KeV)	Int. %	Energy (KeV)	Int. %	Energy (KeV)	Int. %
230	0·3	7·12	230	0·3	..	..	230	2	210	0·8
420 ± 4	0·9	7·41	400	1·0	393	0·76	412	5	420 ± 2	3·7
..	..	..	..	..	..	..	869	9	690 ± 30	6·5
1771 ± 2	47·5	9·0	1760	74	1771	47·6	1756	37	1779 ± 30	33·4
1845 ± 2	52·0	9·1	1840	25	1854	51·6	1839	47	1859 ± 3	48·8

ACKNOWLEDGEMENTS

One of the authors (Ramaniah) is thankful to CSIR for the financial assistance through Post-Doctoral Fellowship.

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1. Marklund, I., Nooijen, B. V. and Grabowski, Z., *Nucl. Phys.*, 1960, **15**, 533.
2. Cork, J. M., Brice, M. K., Helmer, R. G. and Woods, Jr. R. M., *Phys. Rev.*, 1958, **110**, 526.
3. Burson, S. B., Goudsmit, P. F. A. and Konejn, J., *Ibid.*, 1967, 1958, 1161.
4. Lakshminarayana, V. and Subba Rao, K., Private communication, 1973.
5. Cline, J. E., Yates, E. C. and Turk, E. H., *Nucl. Phy.*, 1962, **30**, 154.
6. Nagarajan, T., Ravindranath, M. and Reddy, K. V., *Nucl. Instr. Meth.*, 1969, **67**, 77.
7. Venkata Ramaniah, K., *Ph.D. Thesis*, Andhra University, 1975.
8. Way, K., *Nucl. Data Sheets*, 1964, **6**.
9. Sunyar, A. *W.*, *Phys. Rev.*, 1954, **93**, 1345.
10. Graham, Wolfson and Clark, *Phys. Rev.*, 1955, **98**, 1173 A.
11. Funke, L., Hohmuth, K., Jungclaussen, H., Kaun, K., Muller, G. and Sodan, H,, *Werner Kernenergle*, 1963, **6**, 152.

INDIAN SOCIETY OF CELL BIOLOGY

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ELECTRONIC ABSORPTION SPECTRA OF CHARGE TRANSFER INTERACTIONS INVOLVING SULFUR DONORS

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ABSTRACT

The charge transfer interactions of a series of differently substituted thioureas with iodine in dichloromethane have been studied by electronic absorption spectroscopy. The interaction of one thiourea, di-*t*-butyl thiourea with a strong acceptor, iodine monochloride has also been studied. The spectroscopic and thermodynamic data indicate that thioureas are strong electron donors. Phenyl isothiocyanate is, comparatively, a weaker electron donor.

THE charge transfer complexes formed by *n* donors with σ acceptors are highly stable[1,2] whose enthalpies vary from 3 to 12 kcal mole^{-1}. Since sulfur atom has low ionization potential, it is expected that it will be a better *n*-electron donor than nitrogen atom. It will be interesting to investigate the lone pair donating ability of sulfur atom in comparison with that of nitrogen atom. For this purpose the compounds containing thiocarbonyl (C=S) group with varying substituents would be ideal. A few studies by Drago and coworkers[3] and Rao and coworkers[4,5] with I_2 as electron acceptor have shown that thiourea derivatives are stronger electron donors than amines. Another aspect of interest is that in thiourea derivatives, it is possible that the interaction may occur through nitrogen or sulfur atoms. In view of the exceptionally strong electron donating ability of substituted thioureas, it was considered interesting to examine the interaction of some more thioureas containing widely varying substituents with molecular iodine. Since iodine monochloride is considered to be a better electron acceptor than iodine, the study of the interaction of a few thioureas with iodine monochloride would be of value. The interaction of phenyl isothiocyanate with iodine has also been studied.

MATERIALS AND METHODS

The thioureas were prepared by the reaction between corresponding amines and isothiocyanate and purified[6]. All the solvents and other chemicals were purified by standard methods[7]. The electronic absorption spectra were recorded employing a Perkin Elmer Hitachi 139 Spectrophotometer equipped with a temperature regulated cell holder. Matched quartz cells (stoppered) of 1 cm path length were used. Equilibrium constants (K) were calculated from modified Benesi–Hildebrand equation[8]. The uncertainty in K is less than 10% and the uncertainty in $-\Delta H°$ is $\pm$ 0·5 kcal mole^{-1}.

The approximate oscillator strengths (*f*) for the shifted bands and for pure acceptors were calculated from the relation

$$f = 4 \cdot 319 \times 10^{-9} (\epsilon_m \cdot \Delta\nu_{1/2})$$

where ϵ_m is molecular extinction coefficient at the maxima of the shifted band and $\wedge_{1/2}$ is the width in cm^{-1} of the shifted band at half intensity. The transition dipole moments (D) were calculated from the relation

$$D = 0 \cdot 0958 \left[\frac{\epsilon_m \cdot \Delta\nu_{1/2}}{m}\right]^{1/2}$$

where ϵ_m and $\Delta\nu_{1/2}$ are same as above. ν_m is the frequency of the shifted band maxima in cm^{-1}.

RESULTS AND DISCUSSION

It can be seen from Table I that N-phenyl, *i*-propyl N′-di-*i*-propyl thiourea ($R_1 = C_6H_5$, $R_2 = R_3 = R_4 = i\text{-}C_3H_7$, VII) has the highest equilibrium constant (18,500 l mole^{-1}) and $-\Delta H°$ (14·5 kcal mole^{-1}) with iodine. In general, for the various thioureas, the equilibrium constants vary from 4,400 to 18,000 l mole^{-1} and enthalpies ($-\Delta H°$) vary from 9·6 to 14·5 kcal mole^{-1}. The magnitudes of these values suggest that the substituted thioureas under study are quite a strong electron donors towards the acceptor iodine. The ranges of values obtained for K and $-\Delta H°$ are close to those obtained by Rao and coworkers[4] on simple alkyl substituted thioureas. It can further be seen that in a series of phenyl thioureas, increasing alkyl group substitution, increases the equilibrium constants. Thus, simple phenyl thiourea ($R_1 = C_6H_5$, $R_2 = R_4 = H$, II) has an equilibrium constant of 5,600 l mole^{-1}. The introduction of the one methyl group as in the case of N-phenyl N′-methyl thiourea ($R_1 = C_6H_5$, $R_2 = R_3 = H$, $R_4 = CH_3$, III) increases the equilibrium constant to nearly twice the value (12,800). Introduction of three methyl groups as in N-phenyl, methyl N′-dimethyl thiourea ($R_1 = C_6H_5$, $R_2 = R_3 = R_4 = CH_3$, V) increases the equilibrium constant to 16,270 l mole^{-1}. Similar

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TABLE I

Spectroscopic and thermodynamic data for **Iodine-$R_1R_2NCSN'R_3R_4$** *systems in dichloromethane*

No.	Name of Thiourea	Formula				λ CT (nm)	ϵ	*f*
		R_1	R_2	R_3	R_0			
I	Thiourea(a)	H	H	H	H	298	52,000	1·20
II	Phenyl thiourea	C_6H_5	H	H	H	302	1,800	$1\cdot90\times10^{-2}$
III	N-phenyt, N′-methyl thiourea	C_6H_5	H	H	CH_3	300	1,560	$1\cdot60\times10^{-2}$
IV	N-phenyl, N′-diethyl thiourea	C_6H_5	H	C_2H_3	C_5H_5	304	1,450	$1\cdot90\ \times10^{-2}$
V	N-phenyl, methyl, N′-dimethyl thiourea	C_6H_5	CH_3	CH_2	CH_2	310	1,740	2.18×10^{-2}
VI	N-phenyl. ethyl. N′-diethyl thiourea	C_6H_5	C_2A_5	C_2H_5	C_2H_5	312	1,680	$1\cdot83\times10^{-2}$
VII	N-pheny[, *i*-propyl, N′-di-*i*-propyl thiourea	C_6H_5	i-C_H_7	i-C_3H_7	i-C_3H_7	306	1,800	$2\cdot05\times10^{-2}$
VIII	N-phenyl, *n*-butyl, N′-di-*n*-butyl thiourea	C_6H_5	n-C_4H_9	n-C_4H_9	n-C_4H_9	308	1,600	$1\cdot90\times10^{\ 2}$
IX	N-phenyl, N′-bromo-pyridyl thiourea	C_6H_5	H	H	Br-pyridyl	332	1,902	$7\cdot14\times10^{-2}$
X	N-*p*-tolyl, N′-pyridyl thiourea	*p*-tolyl	H	H	Pyridyl	320	3,050	$8\cdot50\times10^{-2}$
XI	Phenyl isothiocyanate	..	..	..	..	310	..	..

No.	Name of Thiourea	D (Debye)	K (litre mole^{-1}) (30°)	—△H° (kcal mole^{-1})	—△S° (e.u)	b^2/a^2
I	Thiourea(a)	8·75	4,400	9·6	14·7	0·10
II	Phenyl thiourea	1·60	5,600	11·2	19·8	0·12
III	N-phenyl, N′-methyl thiourea	1·55	12,800	14·3	28·4	0·15
IV	N-phenyl, N′-diethyl thiourea	1.23	14,300	13·0	23·9	0·14
V	N-phenyl, methyl, N′-dimethyl thiourea	1·15	16,270	13•2	24·3	0·15
VI	N-phenyl, ethyl, N-diethyl thiourea	1·21	16,420	14·0	26·9	0·15
VII	N-phenyl, *i*-propyl, N′-di-*i*-propyl thiourea	0·92	18,500	14·5	28·3	0·16
VIII	N-phenyl, *n*-butyl, N′-di-*n*-butyl thiourea	0·93	17,650	14·0	26·8	0·15
IX	N-phenyl, N′-bromo-pyridyl thiourea	2·25	14,230	12·5	22·3	0·15
X	N-*p*-tolyl, N′-pyridyl thiourea	2·25	16,190	13·3	24·6	0.15
XI	Phenyl isothiocyanate	..	53·0	3·6	4·0	0·04

thing happens with ethyl, *i*-propyl and *n*-butyl substituents. These variations are clearly due to the inductive effects of the alkyl substituents. The replacement of one of the hydrogens in phenyl thiourea by a bromoperidyl group ($R_1 = C_6H_5$, $R_2 = R_3 = H$, R_4 = Br-Pyridyl, IX) enhances the equilibrium constant and $- \Delta H°$. In the case of N-*p*-tolyl, N′-pyridyl thiourea (R_1 = *p*-tolyl, $R_2 = R_3 = H$, R_4 = pyridyl, X), where we have bulky substituents on either side, the eqilibrium constant is 16,190 l $mole^{-1}$ and $- \Delta H°$ is 13·3 kcal $mole^{-1}$. Steric effects do not seem to play a major role in these interactions.

In the infrared spectra of all the thioureas, the mixed vibration bands with appreciable contribution of the C=S stretching vibration (~ 1070 cm^{-1}) showed lowering of frequency and increases in intensity. This shows that the interaction of iodine occurs through the sulfur atom and not nitrogen.

The oscillator strength of CT bands are appreciable suggesting thereby stronger mode of interaction between iodine and alkyl thioureas. The b^2/a^2 values are a little higher than those for simple alkyl substituted thioureas.

Next, the interaction of iodine monochloride with a few alkyl substituted thioureas have been attempted in dichloromethane as solvent. Unfortunately, thiourea and di-*t*-butyl thiourea gave precipitates with iodine monochloride in dichloromethane solvent system. Di-*i*-propyl thiourea did not give any precipitate and hence this system was investigated in detail. Here the concentration of di-*i*-propyl thiourea was kept constant and the concentration of iodine monochloride varied. The charge transfer transition was located around 305 nm, whose intensity increases with the increase in iodine monochloride concentration. The band around 260 nm characteristic of C=S group in the thiourea, decreased with an increase in the concentration of iodine monochloride. An isobestic point was clearly seen around 270 nm. The equilibrium constant and enthalpy were found to be 18,000 l $mole^{-1}$ and 13·2 kcal $mole^{-1}$ respectively. The values are higher than those with iodine. This can be explained as due to high electron affinity of iodine monochloride.

Next, the interaction of iodine with phenyl isothiocyanate containing —N=C=S group was investigated in dichloromethane. A charge transfer transition was observed around 310 nm and the equilibrium constant and enthalpy are estimated to be 53·0 l $mole^{-1}$ and 3·6 kcal $mole^{-1}$ respectively. This shows that the isothiocyanate group is not effective in charge transfer interactions. This is possibly due to the mesomeric interaction of the nitrogen with the thiocarbonyl group.

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1. Foster, R., *Organic Charge Transfer Complexes*, Academic Press, New York, 1969.
2. Rao, C. N. R., Bhat, S. N. and Dwivedi, P. C., *Appl. Spec. Revs.*, 1972, 5, 1.
3. Drago, R. S., Niedzielski, R. J. and Middaugh, R. L., *J. Amer. Chem. Soc.*, 1964, 86, 1694.
4. Bhaskar, K. R., Gosavi, R. K. and Rao, C. N. R., *Trans. Farad. Soc.*, 1966, 62, 29.
5. —, Bhat, S. N., Murthy, A. S. N. and Rao, C. N. R., *Ibid.*, 1966, 62, 788.
6. Vogel, A. I., *Practical Organic Chemistry*, E.L.B.S. and Longmans Green and Co. Ltd., London, 1962.
7. Weissberger, A., *Technique of Organic Chemistry*, Vol. VII : *Organic Solvents, Interscience*, New York, 1955.
8. Benesi, H. A and Hildebrand, J. H., *J. Amer. Chem. Soc.*, 1949, 71, 2703.

AWARD OF RESEARCH DEGREE

Tamil Nadu Agricultural University, Coimbatore, has awarded the Ph.D. degree in Agriculture to Shri G. V. Kothandaraman, and Shri M. R. Iruthaya Raj.

Karnatak University, Dharwar, has awarded the Ph.D. degree in Chemistry to Shri Veerappa Basavanneppa Jigajinni ; Shri Bhimagonda Revappa Havinale.

The University of Mysore has awarded the Ph.D. degree in Chemistry to Shri P. G. Ramappa.

Osmania University, Hyderabad, has awarded the Ph.D. degree in Chemistry to Smt. Y. Uma Devi.

SECOND MOMENT ANALYSIS OF THE WIDE-LINE NMR SPECTRA OF THREONINE AND ITS ISOMERS

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ABSTRACT

The three crystalline forms of the isomers of threonine arising from the presence of two asymmetric carbon atoms have been analysed by wide-line NMR. A second moment analysis shows interesting configurational variations.

1. INTRODUCTION

RECENTLY we have been interested in the wide-line PMR studies of the amino acids and related compounds. Such a study would be an important adjunct to the X-ray[1,2] and neutron diffraction data[3] and would enable one to correlate and or complement the structural and conformational information from the different sources. Not much of wide-line NMR work has been done on such compounds, although organic and inorganic substances have been studied as shown by a recent survey[4].

This paper deals with the results of the second moment analysis of the amino acid, threonine and its isomers. Threonine is an amino acid containing two asymmetric carbon atoms and therefore has a number of isomers. In fact, a number of different crystalline forms involving threonine and allothreonine have been X-ray analysed in this laboratory[5-8] and compareded[9,10] with structural and conformational information available for other threonine derivatives in the literature.

The close isomorphism of L-threonine and L-allothreonine as determined by X-ray method, led to the possibility of growing a mixed crystal of the two isomers and the X-ray analysis has been reported on a 1 : 1 mixed crystal system[8]. Evidence of equal proportion of the two isomers from the X-ray data is reasonably convincing, however, it was thought worthwhile to examine these systems by PMR technique since such a study on organic molecules does not appear to have been reported in the literature.

2. EXPERIMENTAL

L-threonine and L-allothreonine were obtained from Produits Chimiques—Department of Pharmaceutique, Paris and Nutritional Biochemicals Corporation, Cleveland, Ohio, U.S.A., respectively. The mixed crystal was prepared by the method described elsewhere[8].

The proton magnetic resonance spectra of the polycrystalline samples were recorded at room temperature with a wide-line NMR spectrometer constructed in this laboratory, employing 215 Hz magnetic field modulation and phase sensitive detection. The radio frequency was 11·69 MHz. The audio frequency modulation was kept at about 0·3 G, and the radio frequency amplitude was kept below the saturation level. The line-widths were measured as separation in gauss of points of maximum and minimum slope of the derivative spectra. The experimental second moments for each compound were obtained as a mean from a dozen derivative tracings by numerical integration and were corrected for modulation broadening by standard procedures.[11] All the computations were performed on an IBM–1130 digital computer. A typical derivative tracing of the PMR signal in one of these compounds is shown in Fig. 1.

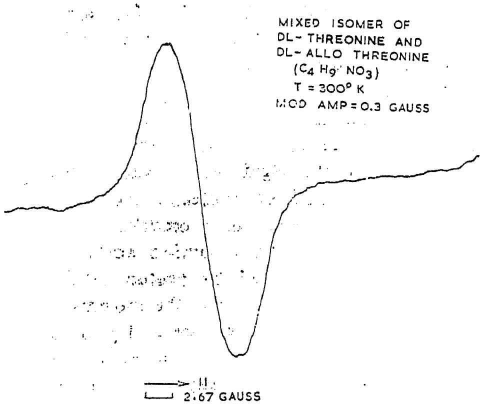

FIG. 1. A typical derivative tracing of PMR Signal in polycrystalline mixed crystal of L-Threonine and L-Allothreonine taken at room temperature.

3. DISCUSSION

Table I shows the second moments deduced from the observed spectra for the three cases. The rigid lattice values of second moment were calculated using Van Vleck's formula for a powdered sample.[12]

$$M_2 = (3/5)\gamma_1^2\hbar^2 I(I+1) N^{-1} \sum_{j>k} r_{jk}^{-6} \qquad (1)$$

where the symbols have their usual meaning and these are also given in Table I. The contribution to the second moment due to magnetic nuclei other than

TABLE I

Second Moment Data for Threonine and its Isomers

Compound	Room temperature second moment (G^2)	Theoretical second moment for a rigid lattice (G^2) Intra	Inter	Total
L-Threonine	13·28 (0·77)	22·40	11·64	34·04
L-Allothreonine	8·54 (0·26)	24·25	12·31	36·56
Mixed crystal of L-Threonine and L-Allothreonine	11·35 (0·23)	24·24	11·57	35·81

Note; Values in porentheses denote standard deviation in experimental values.

protons has been found to be negligible. The sum in (1) is taken over all proton-proton distances less than 10·0 Å, The contribution for $r > 10 \cdot 0$ Å was estimated to be 0·09 G^2 by integration over a continuous distribution of protons. For L-threonine proton coordinates from neutron diffraction study has been used[13], while geometrically fixed positions of hydrogens have been used for L-allothreonine and the mixed crystal, since no neutron diffraction data are available for these. The contribution to the second moment from within a molecule (intra) and neighbouring molecules (inter) is listed separately in Table I. It is seen from Table I that the mean square width (second moment) for the three compounds is very much reduced as compared to the rigid lattice value. This must be due to some kind of moleculr motion in these structures. The presence of molecular motions due to NH_3 and CH_3 groups in amino acids have been identified and characterised by proton NMR relaxation[14] and C_w[15] studies. Since the moleclar motions at room temperature for the pure and allo forms are different, as evinced by different values of the second moments, it is reasonable to expect that a 1 : 1 mixed crystal would give a value of second moment which is in between those of the pure and allo forms. Our experimental value of the second moment for the mixed crystal (11·35 G^2) lies between those of the pure (13·28 G^2) and allo (8·54 G^2) forms. This is in accordance with the earlier X-ray work done in our laboratory.[8] Since these structures have very close cell dimensions, space group and packing, in particular same hydrogen bonding scheme,[10] the intermolecular contribution to the second moment which takes into account the molecular environment is practically the same. Thus the intramolecular contribution reflecets the change in configuration of protons at C^β atom. A detailed study of these compounds and other amino acids at low temperatures is in progress and the results will be reported in due courcse.

ACKNOWLEDGEMENT

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1. Ramachandran, G. N. and Lakshminarayanan, A. V., *Biopolymers*, 1966, **4** 495.
2. Lakshminarayanan, A. V., Sasisekharan, V. and Ramachandran, G. N., In *Conformation of Biopolymers*, Ramachandran, G. N. (Ed.), Academic Press, London, 1967, **1**, 61.
3. Ramanadham, M. and Chidambaram, R., *Acta Crystallogr.*, 1975, A **31** (supplement), S 179.
4. Ganapathy, S. and Srinivasan, R., *J. Sci. Ind. Res.*, 1975, **34**, 134.
5. Swaminathan, P., *Acta Crystallogr.*, 1975, **B31**, 1608.
6. — and Srinivasan, R., *Ibid* 1975, **B 31**, 217.
7. — and —, *J. Cryst. Mol. Struct.*, 1975, (In Press).
8. — and —, *Ibid.*, 1975 (In Press).
9. Srinivasan, R., Varughese, K. I. and Swaminathan, P., *Acta Crystallogr.*, 1974, **B 30**, 2270.
10. Swaminathan, P. and Srinivasan, R., *Int. J. Peptide and Protein Res.*, 1975 (In Press).
11. Andrew, E. R., *Phys. Rev.*, 1953, **91**, 425.
12. Van Vleck, J. H., *Ibid.*, 1948, **74**, 1168.
13. Ramanadham, M., Sikka, S. K. and Chidambaram, R., *Pramāṇa*, 1973, **1**, 247.
14. Andrew, E. R., Hinshaw, W. S., Hutchins, M. G. and Canepa, P. C., *chem. Phys. Letters*, 1974, **26**, 50.
15. Ratkovic, S., *Ibid.*, 1972, **17**, 623.

DIABETIC ASSOCIATION OF INDIA

The Diabetic Association of India, offers twenty travelling fellowships of Rs. 500 each to the Scientists under 35 years who participate in the IX Congress International Diabetes Federation to be held in New Delhi, from October 31 to November 5, 1976. Details can be had from The Hon. Secretary, Scientific Section, Diabetic Association of India, Maneckji Wadia Building, I Floor, Mahatma Gandhi Road, Bombay 400 023.

CHEMICAL EXAMINATION OF *AMARANTHUS SPINOSUS* LINN.

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ABSTRACT

The petroleum-ether extract of *Amaranthus spinosus* L. on application of chromatographic techniques afforded *n*-alkanes C_{23}–C_{33} and iso (-2-methyl)–alkanes C_{29}–C_{33}; esters : transesterified and resolved into acids C_{18}–C_{32} (as methylesters) and free alcohols C_{20}–C_{26}; aliphatic alcohols C_{10}–C_{32}; sterols (β-sitosterol, stigmasterol, campesterol and cholesterol) and free acids C_4–C_{32} (C_{18} group contains stearic acid, oleic acid and linoleic acid). The free amino acids identified are leusine, valine, tryptophan, alanine and serine in the aqueous ethanolic extract.

AMARANTHUS SPINOSUS LINN. (Amaranthaceae) is an important medicinal plant growing wild locally and used to cure various diseases. The ash of the leaves is used in dyeing and the plant as a whole is used in snake-bite and as fodder[1–2]. The leaves of the plant contains oxalic acid[3] and sugar[4] while hemaglutins[5] and betacyanin[6] are present in seeds. In view of the important medicinal properties of the herb and the fact that almost no systematic work has been done so far, a chemical examination of this herb has been undertaken, the results are presented in this paper.

EXPERIMENTAL

Air-dried and powdered plant material (2 kg) collected from surroundings of Aligarh District (India) was extracted thrice with 15 lit. distilled light petroleum (60–80°) at its boiling temperature, which after removal of the solvent under reduced pressure gave a semi-solid mass (1·18%). This extract was separated into acidic (25·2%) and neutral parts (65·4%) by washing with KOH solution (10%). The purified neutral fraction (15 g) was taken in benzene and subjected to chromatographic separation over alumina (∼ 400 g) and two major fractions were collected by eluting the column with petroleum-ether (60–80^0) and benzene.

Petroleum-ether fraction (4·2 g) on crystallisation from acetone-alcohol yielded a colourless waxy solid (400 mg), m.p. 62–63 C°. It showed two spots on T.L.C. (silica gel-2% $AgNO_3$) and therefore was resolved on a column of slicica gel into two parts. Hydrocarbon fraction was eluted with hexane and the ester fraction with chloroform. The separated portions were further analysed by G.L.C. [Gas chromatography was done on PYE series 104 Chromatograph Model 124 with flame inonisation detectors, two column packed with 3% SE–30 on Gas Chrom Z at programmed temperature 150–250 °C (2° C min^{-1}). For identification of homologues the graphical method was employed[7]. The comparison of the retention values with those of standards was used for identification of the gas chromatographic peaks. For a quantitative evaluation areas under the peaks have been calculated.]

Hydrocarbons form two homologous series. One of the series consists of *n*-alkanes (C_{23}–C_{33}) with maximum occurrence of *n*-hentriacontane (C_{31}) (18·1%), *n*-non-acosane (C_{29}) (15·4%), *n*-heptacosane (C_{27}) (9·0%), *n*-octacosane (C_{28}) (8·3%), *n*-triacontane (C_{30}) (8·0%), *n*-dotriacontane (C_{32}) (4·9%), *n*-tritriacontane (C_{33}) (4·5%) and *n*-hexacosane (C_{26}) (3·9%) while *n*-tricosane (C_{23}), *n*-tetracosane (C_{24}) and *n*-pentacosane (C_{25}) were in traces. The other series consists of iso (= 2-methyl)-alkanes (C_{29}–C_{33}) with a maximum of C_{31} (21·1%), C_{29} (3·4%) and C_{33} (2·1%) while C_{30} and C_{32} were in traces; the structure of 2-methyl ocotacosane (C_{29}) and 2-methyl triacontane (C_{31}) in this series was established by G.L.C. combined with mass spectrum. Odd numbered members were prevalent[8–9] in both the series.

Ester fraction showed characteristic i.r. bands at 1176, 1727 cm^{-1} and was transesterified with methanol (+HCl) and the resulting products were gas chromatographed (as methyl esters of the acid and free alcohols). The series of *n*-acids (C_{18}-C_{32} in the form of methyl esters) contained mainly C_{30} (47·7%), C_{28} (28·9%) C_{32} (8·6%); C_{18}, C_{20}, C_{22}, C_{24} and C_{29} were in small amounts (1·0–3·0%) while C_{19}, C_{21}, D_{22}, C_{25}, C_{26}, C_{27} and C_{31} were in traces. The even numbered members were found to be predominating. The series of *n*-alcohols (C_{20}, C_{22}, C_{24}, C_{26}) appeared in traces.

Benzene eluate (8·5 g) was further chromatographed over alumina (∼ 250 g) and yielded two products.

Alcohols.—Elution with light petroleum-benzene (60:40) gave the alcohol fraction. It exhibited bands at 1050, 3400 (OH) and 715, 725 cm^{-1} $(CH_2)_n$ in i.r. spectrum (KBr phase). When analysed by G.L.C. it gave a series of aliphatic alcohols C_{10}–C_{32}; even numbered members were predominating, with maximum occurrence of C_{28} (45·3%), C_{30} (19·1%), C_{22} (11·6%), C_{32} (9·6%), C_{28} (6·4%) and C_{24} (2·9%) while C_{14}–C_{21} were present in small amounts and other members were in traces. However‘ it is interesting to mention that even numbered members were not prevalent between C_{14}–C_{21} in the same series. Branched chain alcohols were also present in minute amounts.

From this mixture *n*-octacosanol (C_{28}), m.p. 83–84° C (300 mg) (lit.[10] m.p. 83·4 ° C) could only be crystallised from methanol (T.L.C.-homogeneous) and confirmed by preparation of its acetate[11] m.p. 75–76° C.

Sterols.—On further elution with light petroleum benzene (20: 80) the fraction of sterols (3·4 g) was obtained. It gave positive Liebermann-Burchard test and yellow colour with TNM. Repeated crystallisation from ethanol afforded shining crystals (350 mg) m.p. 158–69° C (T.L.C.-homogeneous). The i.r. spectrum of the sterol exhibited peaks at 3410 (OH) and 1610 cm^{-1} ($C = C$). It showed depression in m.p. when mixed with authentic specimen of β-sitosterol and gave different R_f values (Co-T.L.C.). The crystalline sterol yielded its acetate m.p. 173° C (ν_{max}^{KBr} 1730, 1245 cm.$^{-1}$) and benzoate m.p. 185–86 °C G.L.C. analysis of the product indicated it to be a mixture of β-sitosterol (83·0%), stigmasterol (12·3%) campesterol (2·9%) and cholesterol (1·6%). It is worth mentioning that the presence of cholesterol is a unique finding in a lower plant (herb).

Acids.—The acidic fraction (2·2 g) was subjected to chromatographic purification over silica gel (~ 80 g). Elution with light petroleium-benzene (1:3) afforded a colourless waxy substance (330 mg) m.p. 78–81 °C and showed peaks in i.r. spectrum (KBr) at 718, 728 $(CH_2)_n$; 1710 (CO); 2850 and 2920 cm^{-1} (CH). T.L.C. indicated it to be a mixture which could not be resolved further even by repeated column chromatography. This product on G.L.C. analysis (after esterification) represented a series of acids C_4–C_{32} with C_{16} (27·5%), C_{18} (17·2%), C_{28} (15·9%), C_{30} (10·7%), C_{24} (9·8%), C_{22} (6·0%), C_{26} (6·1%) and C_{23} (2·5%) while the other members were found in traces. C_{18} group contained saturated and unsaturated acids and was further analysed by G.L.C. (3% diethyleneglycol succinate, at 140″C on polar phase) which indicated it to be a mixture of stearic acid (32·6%), oleic acid (52·6%) and linoleic acid (14·7%). Branched chain acids were also present in minute amounts.

Aqueous ethanolic extract.—The defatted plant material was also extracted with aqueous ethanol (1:1) at its boiling temperature and concentrated *in vacuo*. A dark brown syrupy liquid was obtained which showed the presence of amino acids. The concentrated extract was extracted with chloroform to remove colouring matter. Descending paper chromatography[8,12] (Whatman No. 1) of the aqueous alcoholic extract in *n*-butanol : acetic acid : water (4:1:5, v/v organic layer) indicated the presence of leucine, valine, tryptophan, alanine and serine (when sprayed with ninhydrin 0·1% acetone) as free amino-acids and confirmed by direct comparison with their respective reference samples.

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1. Chopra, R. N., Nayar, S. L. and Chopra, I. C., *Glossary of Indian Medicinal Plants*, (CSIR, New Delhi), 1956, p. 15.
2. Kirtikar, K. R. and Basu, B. D., *The Indian Medicinal Plants*, 1918, **2**, 1058.
3. Balasubramanian, S. C., Narsinga Rao, B. S., Ramanmurthi, P. S. V. and Shenolikar, I. S., *Indian J. Med. Res.*, 1962, **50**, 779.
4. Shankar, J. V. and Bharucha, F. R., *Sci. and Cult.*, 1957, **23**, 193.
5. Osman, H. G. and Jwamny, E. W., *J. Chem. U.A.R.*, 1963, **6** (2), 191.
6. Piatteli, M. and Minale, L., *Phytochemistry*, 1964, 3 (5), 547.
7. James, A. T. and Martin, A. J. P., *Biochem. J.*, 1952, **50**, 679.
8. Behari, M. and Andhiwal, C. K., *Ind. J. Chem.*, 1975, **13** (6), 639.
9. Gupta, G. S. and Behari, *M., J. Ind. Chem., Soc.*, 1972, **49**, 317.
10. Coffey, S., *Rodd's Chemistry of Carbon Compounds*, Vol. I B, 2nd Ed., Elsevier Publishing Company, New York, 1965, p. 33.
11. Heilbron, I. M. and Bunbury, H. M., *Dictionry of Organic Compounds*, Vol. IV, 2nd Ed., Oxford University Press, 153, p. 2.
12. Tandon, S. P., Tiwari, K. P. and Saxena, V. K., *Proc. Nat. Acad. Sci. India*, 1970, Sec. A, **40**, 217.

STUDIES ON LANTHANIDE MIXED COMPLEXES

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MIXED chelates of lanthanide ions are comparatively of recent origin. Several lanthanide diketone mixed complexes were reported[1]. The ligands used were either picolinic acid, quanalidinic acid or dipyridyl. Hart and Laming[2] prepared mixed samarium complex with 1, 10 phenanthroline and acetylacetone. Rohatgi and Sen Gupta[3] have extended these studies further and prepared several lanthanide mixed complexes of the type Ln $(\text{Phen})_2$ $(\text{Sal})_3$, where Phen = 1, 0, phenanthroline and Sal — salicylate.

This communication details the preparation and spectral studies of Y, La, Pr, Nd, Sm, Gd, Dy mixed chelates using dimethylglyoxime as primary ligand and bipyridyl and anthranilic acid as secondary ligands. The thermal analysis of neodymium simple and mixed complexes is also reported.

The simple lanthanide dimethylglyoximates were prepared by adopting the procedure of Rao *et al.*[4]. The mixed complexes were prepared adopting the procedure of Vasireddi[5]. In all the mixed complexes the excess ligand was washed with ether. The complexes were vacuum dried for 48 hours and analysed for metal, nitrogen and anion. The analytical data indicated that each lanthanide ion is attached to one mole of dimethylglyoxime and two moles of anthranilic acid and in bipyridyl mixed complexes each lanthanide ion is attached to one mole of dimethylglyoxime and one mole of bipyridyl. Analysis of one lanthanide complex in each case is given below :

$\text{Nd(DMG)}_1\ (\text{AA})_2\ \text{Cl}\ 2\text{H}_2\text{O}$

% M found 23·67 calc. 23·89 ; % N. found 9·15 calc. 9·27 ; % Cl found 5·74 calc. 5·88

$\text{Nd}\ (\text{DMG})_1\ (\text{Bipy})_1\ \text{Cl}_3\ 2\text{H}_2\text{O}$

%M found 26·25 calc. 26·67 ; % N. found 10·12 calc. 10·35 ; % Cl found 19·20 calc. 19·69

Where DMG = dimethylglyoxime ; AA = anthranilic acid ion and BiPy = 2, 2′ BiPyridyl.

RESULTS AND DISCUSSION

Observations in the ultraviolet region are normally subject to two limitations : (*i*) the sparingly soluble nature of complexes in solvents transparent at this region, (*ii*) and the high absorption of the ligands compared to the metal ions. Hence one can look for band shifts and intensity alterations of the ligand alone. In some cases the total disappearance of the ligand bands is also observed. In this investigation in the several mixed complexes only two band maxima are located. The 250 nm band of anthranilic acid is not located in the mixed complexes. In the dimethylglyoxime bipyridyl mixed complexes the characteristic band of dimethyl glyoxime was not located. However, there is a reasonable intensification of absorption in all the complexes. The log ϵ values at different absorption maxima of several lanthanide mixed complexes investigated are in the range 3·90–4·95.

A study of the visible spectra of Pr, Nd, Sm complexes indicates that the characteristic bands of Pr^{3+} ion are located with intensification of absorption in its dimethyl glyoxime-anthranilate. It is also observed that the 485 nm band is split into two inflections and located at 475 nm (log $\epsilon = 1·03$) and 485 nm (log $\epsilon = 0·94$). In the dimethylglyoxime bipyridyl mixed complex only two bands are located at 450 nm (log $\epsilon = 1·50$) and 487 nm (log $\epsilon = 1·74$). In neodymium dimethylglyoxime-anthranilate only two bands are located at 527 nm (log $\epsilon = 0·93$) and 487 nm (log $\epsilon = 1·07$) whereas in its bipyridyl mixed complex only one band is located at 585 nm (log $\epsilon = 1·24$). The characteristic Sm^{3+} ion band is located only in the dimethylglyoxime bipyridyl mixed complex at 405 nm (log $\epsilon = 1·02$).

In a study of the infrared spectra of dimethyl glyoxime anthranilates and bipyridyl mixed complexes shifts in the O–H stretch region and N–H stretch region have been observed. The shift of N–H str. is normally to a lower frequency and it indicates an N→M dative bond. Such a lowering was observed by Rao *et al.*[6], in simple lanthanide anthranilates. Three bands in the region 1510–1625 cm^{-1} were observed in the anthranilic acid mixed complexes and the two higher frequency peaks could be attributed to C=C and the third to the asymmetric COO^- stretching vibration[7].

The characteristic bipyridyl bands around 760 and 745 cm^{-1} which are due to the out of plane bending of the identical groups of four hydrogens are either split into two inflections or shifted in their position in the mixed complexes. The spectral data of Pr and Nd mixed complexes is detailed in Table I.

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TABLE I

Infrared absorption bands (cm^{-1}) of Praseodymium and Neodymium mixed complexes

DMG	AA	Pr (III) complex	Nd (III) complex	Assignment
	3700 sl			
		3650 sh		OH str.
3400 b	3550 bl	3350 bl	3300 3200 bs	N—H str of anthranilic acid
	1675 sm			C=O str.
1620 bl	1625 sh			C=N str.
	1600 sm	1610 sm	1610 ss	
	1580 sh	1575 sl	1575 sm	Asymmetric CCO^- str.
		1550 sl	1510 ss	
		985 sm	980 sm	N—O str.
978 ss				

DMG	Bipy	Pr (III) complex	Nd (III) complex	Assignment
3400 b		3300 bm	3300 bs	O H str.
	3050 low			
1620 bl	1600 ss	1625 sh	1625 sh	C=N str.
	1580 ss	1600 ss	1600 ss	
		1580 bm	1590 sm	BiPyridyl bands
		1500 sm	1500 sm	
	1470 ss	1480 sm	1480 ss	BiPyridyl bands
		1440 ss	1445 ss	
	1260 ss	1245 sm	1250 sl	Orthosubstituted Pyridine ring
	1000 sm	985 sm	990 sm	
978 ss				N—O str.
	760 bs	765 ss	770 ss	Out of plane bending of ring hydrogens
	745 sm	740 ss	740 ss	
		715 l	715 sl	

DMG = Dymethylglyoxime. AA = Anthranilic acid ion. BiPy = 2. 2′ BiPyridyl.

The thermal analysis of simple neodymium, dimethylglyoximate and the mixed complexes indicated dehydration around 160° C. The organic part is eliminated in the mixed complexes in stages. Both in the simple and mixed complexes there is an intermediate halide formation and this can be postulated from weight loss calculations. Such a halide formation was also postulated in a study of thermal analysis of transitional metal pyridine complexes[8] and also in the disprosium dimethyl glyoximate[9]. The sesquioxide is formed between 600–900° C in all the complexes.

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---

1. Dutt, N. K. and Upadhaya, S., *J. Inorg. Nucl. Chem.*, 1967, 29, 1368.
2. Hart, J. A. and Laming, F. P., *Ibid.*, 1964, 26, 519.
3. Rohatgi, K. K. and Sen Gupta, S. K., *Ibid.*, 1970, 32, 2247.
4. Galgali, V. H., Ramachandra Rao, V. and Khanolkar, D. D., *Ind. J. Chem.*, 1969, 7, 825.
5. Vasireddi, S. P., "Studies on rare earth complexes," *Ph.D. Thesis*, Nagpur University, 1974, p. 103.
6. Deshpande, Y. H. and Ramachandra Rao, V., *Ind. J. Chem.*, 1969, 7, 1051.
7. Hill, A. G. and Curran, C., *J. Phys. Chem.*, 1966, 64, 1519.
8. Brown, H., *Proc. Soc. Analyst. Chem.*, 1971, 8 (8), 169.
9. Vasireddi, S. P., *Ph.D. Thesis*, Nagpur University, 1974, p. 375.

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ISOLATION AND INSULATION OF SPECIAL CELLS

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IN unicellular green motile walled algae such as *Chlamydomonas*, during reproduction or cell division, the entire protoplast withdraws sooner or later from the surrounding cell wall. The connections between the external flagella and the internal organelles are severed. The protoplast divides longitudinally. This severance of connections with the external flagella is more obvious when the plane of division is at right angles to the longitudinal axis, and is preceded by a rotation of the entire protoplast by 90°. In some species of *Haematococcus*, where the flagella are not dropped and the rotation of the protoplast is not obvious, careful studies have revealed that the neuromotor apparatus gets disjuncted inside the protoplast at the base of the flagellum and instead of the whole protoplast rotating, there is a rotation of the internal organelles by 90° so that the cell division which appears tranverse is really longitudinal *vis-a-vis* the internal organelle arrangement and its symmetry (Pocock, 1960). What is more interesting is that even in multicellular algae, a withdrawal of the contents from their walls is very common or universal during reproduction. It is often described as a method of eliminating sap vacuoles and the other functional impulses, if any, are missed.

Similar rotation of protoplasts are seen in *Prasiola* during reproduction when areas of the monostromatic thallus becomes polystromatic. The cell division is longitudinal to the symmetry of the protoplast and at 90° to the normal plane of division (Friedmann, 1959). Such thalli, often described as parenchymatous, must be considered as making really a palmelloid organization, an ordered or unordered aggregate of individual cells. The same holds good in the case of some ulotrichalen genera. These lack a fixed polarity, morphologically speaking, and probably behave like palmelloid organisms. It is not surprising that in these cases the life-cycle involves unicellular growing somatic phases as against resting conditions. In other ulotrichalen and chaetophoralen algae protoplasmic connections are known and in these also the protoplasts withdraw from the cell walls before the formation of swarmers or reproductive elements. The protoplasmic connections between sister cells probably function as channels for transport of metabolites and messages and an expression of polarity, controlling activities of the plant body and its contained cells. During reproduction the contents of a differentiating cell contract from the wall all around, thus breaking off the protoplasmic connections with the neighbouring cells and escaping from the restrictive influences or effects from their presence. The fact that internal changes in the location of organelles, especially the nucleus needed for further development, follows such a break from the constraints placed on it, by the presence of protoplasmic connections is significant. The purposiveness of such a withdrawal seems to be more evident when we observe that in some cases the protoplasts of such cells may refill the cell cavity. Such an isolation may sometimes be postponed till the time of shift in the position of the nucleus in male and female cells preparatory to flagellation or fertilization.

In many cases, the withdrawal of cell contents from the cell wall is accompanied by material secretion around the protoplast, a feature seen in some genera even in asexually formed spores or parthenospores or azygospores. While there is a small difference of opinion on the timing of secretion of this material in which it becomes encapsulated as such, most people consider it at least simultaneous with the withdrawal of the protoplast from the wall. In some algae the motile reproductive elements remain enclosed in a thin vesicle for some time or until sometime even after release from the parent cell. Such secretions may insulate as it were, the already directed cell differentiation and thus lead to an ordered progress of a differentiating cell against any further influence or changes detrimental to the differentiation. What functions do these and other secretions play, in generally screening off the differentiated cell after being charged with a function from further directional influence, which may ensue by their connection with the neighbouring cells ? This is not to minimize or negate other functions these may serve. Details of similar mechanisms that may exist in palmelloid aggregates or pseudoparenchymatous tissue of cells are not known.

While this is the case in autochthonously generated process of cell differentiation, allochthonously generated impulses meddling with the structure and organization also simulate these developments. We cite only two diverse examples. When cells of a prothallus are subjected to nonlethal plasmolysis the protoplasts withdraw from the walls and each of them behaves as an independent unit and forms a new plant (Nakazawa, 1963). From recent studies of the nucleus it is becoming more and more evident that the nucleus is variously anchored as as it were, to other cell organelles such as the chloroplast or pyrenoid or may be held in position by an array of microtubules. Any disturbance of this, brought about artificially, not only disturbs or dislodges the nucleus but may set in motion a chain of events or developments resembling one or the other aspects referred to above. For instance, plasmolysis may result. It is not surprising that such a plasmolyzed cell functions as a reproductive cell and forms a new plant (Walker *et al.*, 1975). This also explains the totipotency of single undifferentiated cells such as parenchyma cells when studied in isolation, as these are not subjected to directional influences as may be present in a well-knit tissue (Steward, 1968, p. 467). It is from the same influence a plasmolyzed protoplast escapes by isolation and insulation even while being embeded in a mass of similar cells.

The above examples are instances of isolation and insulation of an ordered differentiating cell in the normal process of an organism without involving any change in the genetic make-up. These same phenomena seem to occur also in differentiating cells which are accompanied by changes in ploidy. In the palmelloid or pseudoparenchymatous genus *Prasiola*, for instance, it is possible for haploid cells derived after meiosis to grow in a diploid environment and form mosaic patches of haploid cells in an apparently different, diploid environment. In the case of angiosperms when a megaspore or microspore mother cell is differentiated, it is separated from the neighbouring cells by a fresh layer of wall material such as callose, before it undergoes reduction division. A comparision of this with the early stages of formation of apomictic embryos reveals the significance of this isolation. This seems to be true of all cells which undergo reduction division, in an otherwise diploid environment or tissue. In some coenocytic conditions, a diploid nucleus becomes selected for reduction division. Such nucleii become enclosed in sporangia or cysts and the reduction division takes place inside these. In others the reducing nuclei, together with a portion of the cytoplasm, become partitioned off from the rest of the protoplast. This does not preclude the possibility of establishment later, of saprophytic or even parasitic connections by fertilized female cells with the parental tissue. In these cases the haploid nucleus of a parasitized cell may even multiply along with diploid nucleus but they appear to have no distinct function and in fact they ultimately become degenerate. How this competitive antagonism between changed nuclei (diploid or haploid) and the unchanged nuclei (haploid or diploid) works out is yet to be fully understood.

Thus it appears that isolation and insulation of a normal differentiating cell or changed cell from further directional messages or harmful or inhibitory metabolites or influences are not only essential for an orderly development of the charged cell, but such an isolation and insulation appears to be necessary in cases of cells, which have a changed genetic component, either normal to the organism or abnormal or even harmful to the organism itself. In the latter group we are inclined to include certain cancer cells. Incomplete isolation and insulation of differentiating, zygotic or embryonal cell within an organism or tissue or environment leads to either a reversal of development or may even prove lethal to the cell. This hypothesis does not apply to a differentiating cell(s) which even when fully differentiated has to remain connected to other cells for its functional activities and insulation is not necessary for differentiating changes. It applies only for such differentiating cells which are independent and need to become independent. Such differentiations can be internally directed and the cells can depend on its surface for select physiological activity as the differentiation may demand. In the establishment of a mutant or changed cell such as cancer cells it may be essential as otherwise influences detrimental to it or defence mechanisms generated by the environment may prove lethal or destabilizing.

Steward (1968, p. 485) has said that what a cell differentiate into is due not so much to what the cells are, as to where they are. The present conception is in a way a complementary (?) one and explains that a cell can become different from the rest of the cells around it or the sister cells even where they are, in other words, 'in it, yet out of it'.

How common are these two phenomena to other living organisms than those that have been dealt with in this short exposition? What is the nature of insulation in these cases and other cases? Does it involve only changes in the plasmalemma or does it involve also extraplasmalemmar secretions or coats or physico-chemical changes? These are the questions that will no doubt be studied and argued in the future.

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1. Friedmann, I., *Ann. Bot. Lond. n.s.*, 1959, 23, 571.
2. Nakazawa, S., *Sci. Rep. Tohuku Univ, 4th ser.*, 1963, 29, 247.
3. Pocock, M. A., *Trans. Roy. Soc. S. Afr.*, 1960, 36, 5.
4. Steward, F. C., *Growth and Organization in Plants.* Addison-Wesley Publ. Co., Reading, Mass., 1968, 564 pp.
5. Walker, D. C., Chi, E. Y., and Neushul, M., *Am. J. Bot.*, 1975, 62 (9), 901.

RELEASE OF EXTRACELLULAR MATTER DURING PHOTOSYNTHESIS BY A *TRICHODESMIUM* BLOOM

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ABSTRACT

Data presented are interpreted as suggesting the release of dissolved ^{14}C labelled organic matter during the photosynthetic fixation of carbon supplied as Na H $^{14}CO_3$ by a natural bloom consisting largely of the blue-green alga, *Trichodesmium.*

INTRODUCTION

It is well established that most phytoplankton cells both in culture and the natural environment release extracellular products during the course of normal photosynthesis (*see* review by Fogg, 1975 *a*)[1]. The occurrence of *Trichodesmium* blooms on the west coast of India have been reported earlier (Qasim[2], Ramamurthy *et al.*[3]). During March to May of 1975 productivity estimations were carried out on a bloom of the blue-green alga *Trichodesmium* which occurred between the 10–20 m depth-zone (1 to 4 km off shore) in the neritic waters off Goa (Devassy *et al.* to be published). This report concerns estimates of the production of extracellular matter during photosynthetic carbon fixation on samples collected during the course of this investigation.

METHODS AND MATERIALS

The bloom was originally sighted in the 10 to 20 m depth-zone off Goa in late February. Regular collections were made thereafter at 5–8 day intervals. Stations were fixed arbitrarily depending on the position and apparent density of the bloom patches. The depth of the *Trichodesmium* patch seldom exceeded 0·5 m. The dominant organism present was *T. erythraeum,* although some *T. thiebutii* was also found in the initial stages of the investigation.

Water samples were collected from the surface and 2 m depths and subsampled into 120 ml light and dark glass bottles. Following inoculation with ^{14}C labelled sodium bicarbonate (4 μCi) they were incubated in a deck incubator in full sunlight. Samples from the 2 m depth were covered with socks of blue net to cut down light to the appropriate intensity of 35% surface irradiance. Light and dark bottles from both depths were filtered in the laboratory using 0·45 μ. Sartorius membrane filters. The period between incubation and filtration did not exceed 1 hour. The samples were kept in the dark at water temperature during the intervening period.

A 50 ml aliquot of the filtrate was strongly aerated at a pH of 4 for 45–60 minutes and, following neutralization to a pH of 10, a 100 μl sample was counted in 10 ml of a dioxan-based scintillation cocktail. Radioactivity on the filters was counted using a similar geometry. Radioactivity determinations in the light filtrates have not been corrected for dark values (*see* Fogg, 1975 *b*)[4].

Chlorophyll *a* and phaeophytin determinations were made spectrophotometrically following the method published in Strickland and Parsons (1968)[5]. Cell counts and other data have been taken from Devassy *et al.* (to be published).

RESULTS AND DISCUSSION

Evidence leading to the hypothesis that this *Trichodesmium* bloom was the result of a convergence rather than a sudden growth of the alga in a discrete water mass has been discussed by Devassy *et al.* (to be published). It is worth reiterating here that whereas nitrate values do not vary significantly from the normal, phosphate values in these samples are in the region of 1–2 μ gatom per litre, almost 4 times higher than the expected average of 0·5 μ gatom per litre at this time of year. This, in conjunction with high phaeophytin, suggests that the bloom may have already passed its peak at the time of investigation. However, particulate fixation of inorganic carbon was 3 to 4 times higher (Table I) than the average of 16·25 mg $Cm^{-3}h^{-1}$ recalculated from Qasim *et al.*[6] 1969, for the Cochin Backwaters and suggest that a substantial part of the population consists of viable cells.

It is possible that the delay in filtration has influenced determinations of radioactivity of both particulate carbon fixation and the production of extracellular material. Dark fixation of inorganic carbon and bacterial uptake of organic carbon in the dark, for example, may lead to an overestimation of inorganic carbon fixation by algae and an underestimation of extracellular production. In

TABLE I

Light bottle measurements of particulate fixation and extracellular material. Production has been calculated according to Watt (1966). Dark extracellular production is presented as per cent fixation of corresponding particulate carbon uptake

					Light bottle values only			Dark values
Date	Station and depth	Chlorophyll *a* mg m^{-3}	Pheophytin mg m^{-3}	% Trichodesmium (cell count)	Partieulate fixation mg C m^{-3} hr^{-1}	Extracellular production mg C m^{-3} hr^{-1}	% Extracellular release	% Extracellular release
5–3–75	St. 1 0m	0·05	0·0	58·9	6·14*	0·0	0·0	0·0
10–3–75	St. 1 0m	0·05	0.0	10·0	3·0 ± 0%	0·0	0·0	0·0
22–3–75	St. 1 0m	0·5	0·9	57·5	46·4 ±30%	2·7 ±16%	5·9	19·57
	2m	0·0	0·0	26	6·6 ±16%	0·98 ±30%	14·85	50
	St. 2 0m	26·7	46·7	75·8	45·26 ±18%	31·3 ±21%	69·2	31·98
	2m	0·59	0·0	57·5	13·9 ±58%	2·7*	19·2	28·1
29–3–75	St. 1 0m	0·0	3·6	0·96	27·7 ±63%	1·8 ±100%	6·5	4·7
	2m	0·0	0·5	0·0	29·6 ±15%	0·2 ± 0%	0·6	75·1
	St. 2 0m	6·0	0·0	92·6	89·96 ±71%	10·6 ± 76%	11·2	51·8
	2m	0·08	0·22	3·45	25·15 ±5%	2·0 ±100%	7·7	..
2–4–75	St. 1 0m	0·0	2·8	37·0	10·7 ±10%	1·2 ±55%	11·4	..
	2m	0·0	7·2	0·0	16·41 ±29%	0·0 ±0%	44·2	100·0
	St. 2 0m	4·1	0·0	99·6	54·2 ±10%	10·9 ±29%	20·2	57·1
	2m	0·9	0·0	18·2	17·4 ±78%	..	..	23·3
7–4–75	St. 1 0m	0·5	0·4	21·7	38·7 ±84%	19·7 ±53%	50·8	..
	2m	0·07	0·09	0·0	15·5 ±51%	5·9 ±22%	38.14	100·0

* Only one sample.

spite of such errors, however, the data presented in Table I suggest that some release of organic matter occurred during photosynthesis in these samples. On the whole, production of extracellular material varies from 0·2 mg m^{-3}hr^{-1} to 31 mg m^{-3}hr^{-1} in the light bottles, which is within a range of 1–69% of the light bottle inorganic carbon fixation by algal material. Even in cases where as much as 99% of the cell population consists of *Trichodesmium* filaments some apparent release of ^{14}C labelled organic matter occurs.

In terms of mg C m^{-3}hr^{-1}, dark bottle release of extracellular material is lower than in the light, although such fixation expressed as a percentage of carbon uptake is higher. A certain amount of difficulty has been experienced in the processing of these samples due to the sticky nature of the clumps of *Trichodesmium* filaments and the mucilaginous material they produce. Perhaps as a result of this standard deviations of both inorganic carbon fixation and extracellular production were high even on samples taken on the same day and depth. However, the overall averages for per cent light and dark bottle release are 18·2 and 37·3% respectively and in spite of the high standard deviations from the samples it seems likely that the differences are real. The higher values of per cent release in dark bottles are a probable result of low inorganic carbon uptake in the dark.

Samples from the surface generally yield higher values of extracellular production in terms of mg C $m^{-3}hr^{-1}$ (Table I) than the samples from the 2m depth when both sets of samples were incubated under simulated *in situ* conditions. This may be a result of both the inhibiting light intensity at the surface ($>$ 500 gm cal $cm^{-2}day^{-1}$) and lower cell concentrations at the 2m depth leading to lower actual production values. Watt (1966)[7] has found that there is an increase in per cent release of extracellular matter with depth associated with the dark fixation of inorganic carbon. Per cent release in these samples, however, shows a great deal of variation depending both on stations and dates of sampling.

While it is known that some blue-green algae liberate nitrogenous products (Fogg, 1952)[8], little information is available on the extracellular products of *Trichodesmium* during photosynthetic carbon fixation either in laboratory cultures or the natural environment. The series of histograms presented in Fig. 1 represent data from those surface samples where cell concentrations were comparatively high on any given day of sampling. Neither particulate carbon fixation nor extracellular production show a correlation with total cell counts. The peaks in both extracellular production and per cent release of organic matter, however, seem to follow a trend similar to that of chlorophyll *a* where the latter values have been corrected for phaeophytin. This appears to suggest that extracellular release in these samples may be related to photosynthetic precursors in *Trichodesmium*, particularly in those cases where the alga is clearly the dominant organism and perhaps responsible for most of the active chlorophyll.

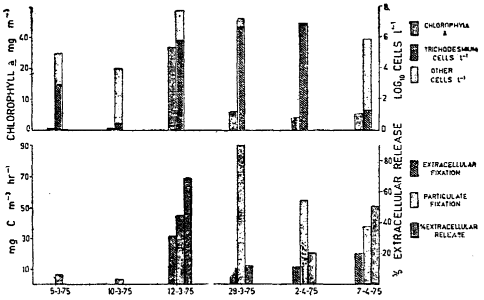

FIG. 1. Histograms representing data from selected stations. Surface light bottle samples only. Particulate fixation has not been corrected for dark uptake.

Samuel *et al.* (1971)[9] found that phytoplankton populations in the Cochin Backwater release extracellular matter as a result of inorganic ^{14}C fixation. Work in this laboratory substantiates these results for natural populations under non-bloom conditions in waters off Goa. It seems possible therefore, that both flagellates and diatoms associated with the *Trichodesmium* bloom are responsible for part of the extracellular production reported here and that this is likely to originate from recently fixed photosynthetic products.

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1. Fogg, G. E., *I. B. P.*, 1975 *a*, 3, 437.
2. Qasim, S. Z., *Deep-Sea Res.*, 1970, 17, 655.
3. Ramamurthy, V. D., Selvakumar, R. A. and Bhargava, R. M. S., *Curr. Sci.*, 1972, **41**, 803.
4. Fogg, G. E., In *Chemical Oceanography* (Eds. Riley, J. P. and G. Skirrow), Acad. Press, 1975 *b*, 2, 386.
5. Strickland, J. D. H. and Parsons, T. R., *Fish. Res. Bd. Canada, Bull.*, 1968, 167, 311.
6. Qasim, S. Z., Wellershans, S., Bhattithiri, P. M. A. and Abidi, S. A. H., *Proc. Ind. Acad. Sci.*, 1969, 69 B, 51.
7. Watt, W. D., *Proc. Roy. Soc.*, 1966, 164 **B**, 521.
8. Fogg, G. E., *Ibid.*, 1952, 139, 372.
9. Samuel, S., Shah, N. M. and Fogg, G. E., *J. Mar. Biol. Assoc.* (U.K.), 1971, 51, 793.

LETTERS TO THE EDITOR

ASSOCIATED KERR METRIC

We have expressed the Kerr solution[1] of Einstein's equations $R_{ik} = 0$ admitting shearfree, geodetic, twisting null rays in the following form[2]:

$$ds^2 = 2(du + k \sin^2 \alpha d\beta)\,dt - [1 - 2\mathrm{M}r/(r^2 + k^2 \cos^2 \alpha)] \times (du + k \sin^2 \alpha d\beta)^2 - (r^2 + k^2 \cos^2 \alpha)(d\alpha^2 + \sin^2 \alpha d\beta^2). \tag{1}$$

Here t is the galilean time, r the radical distance and u is the retarded distance ($u = r + t$) in the Minkowskian background.

While studying a general metric[3] which can describe the above Kerr solution as well as the similar NUT solution[4] of Einstein's equations, it was found that essentially there are two metrics which describe Kerr solution, the line-element (1) above being one of them. As a matter of fact

$$ds^2 = 2(du + y' \sin^2 \alpha d\beta)\,dt - [1 - 2\mathrm{Mr}/(r^2 + y^2)](du + y \sin^2 \alpha d\beta)^2 - (r^2 + y^2) \times (d\alpha^2 + \sin^2 \alpha d\beta^2)$$

$$y = y(z), \qquad z = \cos \alpha, \quad y' = dy / dz. \tag{2}$$

expresses the Kerr solution of Einstein's equations if Y satisfies Legendre's equation of order 1 *viz.*,

$$y''(1 - z^2) - 2zy' + 2y = 0. \tag{3}$$

One solution of (3) is $y = kz = k \cos \alpha = kp_1(z)$, p_1 being Legendre's polynomial of order 1. This solution when used in (2) gives the standard Kerr metric (1). But $y = kQ_1(z)$ also satisfies (3) where Q_1 is the corresponding associated Legendre function[5]. Thus along with the usual Kerr metric (1) we have the Associated Kerr Metric.

$$ds^2 = 2(du + kQ_1' \sin^2 \alpha d\beta)\,dt - [1 - 2\mathrm{M}r/ \times (r^2 + k^2 \mathrm{Q}_1{}^2)](du + k\mathrm{Q}_1' \sin^2 \alpha d\beta)^2 - (r^2 + k^2 \mathrm{Q}_1{}^2)(d\alpha^2 + \sin^2 \alpha d\beta^2). \tag{4}$$

Since the associated Legendre function Q_1 becomes infinite on the axis $\alpha = 0$, the associated Kerr Metric has a singularity along the axis $\alpha = 0$.

It is further found that metrics of the type (2) can represent radiating Kerr and NUT space-times if Y satisfies Legendre's equation of general order n instead of equation (3) which is of order 1. All these results are being reported elsewhere[3].

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1. Kerr, R. P., *Phys. Rev. Lett.*, 1963, **11**, 237.
2. Vaidya, P. C., *Proc. Camb. Phil. Soc.*, 1974, **75**, 383.
3. —, Patel, L. K. and Bhatt, P. B., *A Kerr-NUT Metric* (Under publication).
4. Newman, E., Tamburino, L. and Unti, T., *J. Math. Phys.*, 1963, **4**, 915.
5. Sneddon, I., *Elements af Partial Differential Equations*, McGraw-Hill, 1957, p. 156.

THE CRYSTAL STRUCTURE OF DIGLYCINE CALCIUM CHLORIDE TETRAHYDRATE

The addition compounds and complexes of amino acids with inorganic compounds are of particular interest chemically in view of the coordination of the metal ion and hydrogen bonding, and in biology for their importance and occurrence in living systems. It is also known that some of the glycine compounds have therapeutic values. Hence, a systematic crystallographic study of the addition compounds of glycine, the simplest amino acid, with calcium halides, was taken up. This is a preliminary note reporting the atomic coordinates in the crystal structure of diglycine calcium chloride tetrahydrate.

Single crystals of diglycine calcium chloride tetrahydrate [$(\mathrm{NH_2CH_2COOH})_2\ \mathrm{CaCl_2} \cdot 4\mathrm{H_2O}$] were grown from a saturated aqueous solution containing stoichiometric amounts of glycine and calcium chloride. The crystal data, as given by Jayalakshmi and Vijayan[1], are as follows: $a = 13 \cdot 01$, $b = 6 \cdot 79$, $c = 15 \cdot 38$ Å, $\beta = 91 \cdot 4°$, $\mathrm{V} = 1358 \cdot 6$ Å^3, F.W. $= 333 \cdot 2$ gm, $\mathrm{D_{mea}} = 1 \cdot 63$ gm cm^{-3}, $\mathrm{D_{eal}} = 1 \cdot 63$ gm cm^{-3}, $\mathrm{Z} = 4$, $\mu(\mathrm{CuK}_\alpha) = 77 \cdot 9$ cm^{-1} and the space group is $\mathrm{P2_1/c}$. The density given in the above data was measured by flotation in a mixture of bromoform and carbon tetrachloride.

Three-dimensional intensity data ($h\mathrm{K}l$, $\mathrm{K} = 0$ to 6, $\mu r = 0 \cdot 96$; $hk\mathrm{L}$, $\mathrm{L} = 0$ to 2, $\mu r = 0 \cdot 78$; $0kl$, $\mu r = 0 \cdot 66$) were collected using CuK_α radiation by the multiple film equi-inclination Weissenberg technique. The spot-shape, absorption, Lorentz and polarisation corrections were applied in the usual way. From sharpened and unsharpened three-dimensional Patterson syntheses, as well as from a three-dimensional minimum function, the positions of the calcium atom and the two chlorine atoms were determined.

Thereafter, successive Fourier and difference Fourier syntheses revealed the rest of the structure. With isotropic thermal parameters for all the atoms, least-squares refinement was carried out using the LALS program (originally written by Gantzel, Sparks and Trueblood and modified later by Liminga of Uppsala and also in the University of Madras) on the IBM 370/155 computer, at the Indian Institute of Technology, Madras. For 1773 observed reflections, the residual index at present is 0·18. Further refinement is under progress. The present fractional atomic coordinates are presented in Table I.

TABLE I

Fractional atomic coordinates for diglycine calcium chloride tetrahydrate

Atom	*x*	*y*	*z*
Ca	0·4155	0·1799	0·2467
Cl (1)	0·0315	0·1219	0·1748
Cl (2)	0·8429	0·2217	0·4661
O (11)	0·592	0·023	0·236
O (21)	0·618	0·340	0·268
C (11)	0·646	0·168	0·247
C (21)	0·762	0·135	0·228
N (11)	0·824	0·316	0·261
O (12)	0·260	0·138	0·333
O (22)	0·286	0·260	0·465
C (12)	0·230	0·203	0·405
C (22)	0·112	0·218	0·407
N (12)	0·076	0·312	0·480
O_w (1)	0·269	0·220	0·149
O_w (2)	0·477	0·263	0·105
O_w (3)	0·472	0·160	0·398
O_w (4)	0·623	0·123	−0·007

The calcium atom is coordinated to seven oxygen atoms, three of them belonging to water molecules and the rest to the carboxyl groups of the glycine molecules. The Ca-O distances range from 2·34 to 2·54 Å No chlorine atom coordinates to calcium. The glycine molecules have the normal bond distances and bond angles as found in other similar structures[2]. There are one or two slight abnormalities in these quantities, which will, however, improve with further refinement. The hydrogen bonds are between the nitrogen, oxygen and chlorine atoms. This structure is not isomorphous with the corresponding bromide compound, *viz.*, diglycine calcium bromide tetrahydrate, the crystal structure of which has also been elucidated by us and will be published shortly.

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1. Jayalakshmi, K. and Vijayan, M., *Acta Cryst.*, 1967, **23**, 669.
2. Mohana Rao, J. K. and Viswamitra, M. A., *Ibid.*, 1972, **28 B**, 1484.

FORMATION OF DISULPHUR MONOXIDE BY THE REACTION BETWEEN ELEMENTAL SULPHUR AND OXY COMPOUNDS

IT has been shown that disulphur monoxide is formed along with sulphur dioxide when elemental sulphur is allowed to react at low pressures and elevated temperatures with a limited supply of oxygen or an oxide of a heavy metal like copper or its sulphate.[1–3] In the present communication we report the formation of disulphur monoxide when elemental sulphur reacts at elevated temperatures in vacuum with a variety of oxygen containing compounds like phosphate, arsenate, antimonate, bismuthate, molybolate, tungstate, permanganate, dichromate and acetate.

1 g of the oxysalt was intimately mixed with 10 g of resublimed elemental sulphur and heated in a pyrex glass tube (30 cm × 2 cm) fitted with appropriate stoppers and connected to a vacuum line. The pressure in the system was maintained at $\sim 10^{-3}$ mm of mercury. The gaseous products were condensed in traps cooled in liquid nitrogen. The deposit of a cherry red mass in the form of a ring inside the cold tube indicated the presence of disulphur monoxide as revealed by analysis[4].

Disulphur monoxide was found to be formed during the reaction of elemental sulphur with all the compounds listed in Table I. The temperature at which the reaction began also is given in the table. It is of interest to observe that the hydrated sulphates reacted at comparatively lower temperatures than anhydrous sulphates. It is probably due to the highly reactive surfaces formed *in situ* by the expulsion of water. However, in this case, a part of the disulphur monoxide gets hydrolysed in the cold trap by the water present and the resulting hydrogen sulphide which is also collected in the trap.

The yield of disulphur monoxide is moderate (20–30 mole%) in all cases excepting in the case of magnesium-sulphate in which it is rather low. The maximum yield

TABLE I

Reaction between elemental sulphur and some oxygen containing compounds in vacuum

Compound	Temperature at which reaction starts °C
Manganese sulphate (anhydrous)	650
Manganese sulphate (hydrated)	500
Ferrie sulphate (anhydrous)	500
Ferric sulphate (hydrated)	450
Cobalt sulphate (anhydrous)	600
Cobalt sulphate (hydrated)	500
Nickel sulphate (anhydrous)	500
Nickel sulphate (hydrated)	400
Copper sulphate (anhydrous)	300
Copper sulphate (hydrated)	200
Magnesium sulphate (anhydrous)	750
Magnesium sulphate (hydrated)	650
Sodium molybdate	400
Sodium tungstate	400
Sodium sulphite	500
Sodium carbonate	500
Sodium bismuthate	500
Copper phosphate	500
Copper arsenate	450
Copper antimonate	400
Copper acetate	550
Cobalt molybdate	500
Potassium dichromate	500
Potassium permanganate	500

was in the case of anhydrous copper sulphate (50 mole %). In the case of sulphates of transition metals of the first series the yield of disulphur monoxide decreased in the order Cu > Ni > Fe > Co > Mn. In the case of copper salts the yield decreased in the order sulphate > antimonate > arsenate > phosphate > acetate. With sodium salts, a variation in the anion does not have much effect on the yield of disulphur monoxide.

The yield of disulphur monoxide seems to be related to the ease of reduction of the oxycompounds by sulphur which is evident from the temperature at which the reaction begins in each case,

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---

1. Murthy, A. R. V., *Proc. Indian Acad. Sci.*, 1952, **36 A**, 388.
2. —, *Nature*, 1962, **193**, 773.
3. Nair, K. R., Kutty, T. R. N. and Murthy, A. R. V., *Curr. Sci.*, 1970, **39**, 322.
4. Murthy, A. R. V., Kutty, T. R. N. and Sharma, D. K., *Int. J. of Sulfur Chem.*, 1971, **6 B$_1$** 159.

POLAROGRAPHIC STUDIES OF INDIUM (III) COMPLEX WITH SODIUM OXALATE

Introduction

DICARBOXYLIC acids are well known for their strong co-ordinating capacities. Complexes of oxalic acids with many metals have been reported[1-4]. Deichman[5] has studied indium oxalate complex, over a wide range of oxalate concentrations by conductance and pH measurements. At pH < 4 the formation of $InC_2O_4^+$ has been shown by Dubovenko[6]. White and co-workers[7] have studied indium (III) complex of bioxalate by liquid ion exchange method. In the present communication the authors have reported the results of polarographic studies in indium-oxalate complex.

Experimental

Indium was [$InCl_3$ (fluka)] and all the other reagents were of Analar quality. The solutions were prepared in double-distilled water. The polarograms were obtained on a Tinsley pen recording polarograph. The pH value of each set was determind by a Philips pH meter. All the operations were carried on as 25° C.

Several sets were prepared, keeping the over all potassium chloride, indium chloride and gelatin concentrations at 1M, $6\cdot25 \times 10^{-4}$ M and 0 005% respectively. The ligand (sodium oxalate) concentration was varied from 0·0025 M to 0·0400 M (Table I).

The sets were kept for 24 hours before recording their polarograms. Each set was deoxygenated by passing pure hydrogen.

The drop time the damping, the sensitivity of the instrument and pH values of all the sets were kept at 3 sec/drop, – 3, 10 μa, 6·3 sensitivity—10μa respectively.

Result and Discussion

Reversibility of the waves was tested by plotting $\log [i/(i_d - i)]$ *vs.* applied potential $(E_{0\cdot5})$ Straight lines were obtained with the slope of $0\cdot0591/n$ volts confirming reversibility of the waves[8].

The experimental value for the $E_{0\cdot5}$ of In (III) is $-0\cdot615$ V. Addition of sodium oxalate shifts the half wave potential to a more negative value indicating the formation of complex[9]. The metal ion is reduced to a lower oxidation state while the complex remains dissolved in the solution phase. Further more, the half wave potential is more negative, the smaller the value of K_{instab}.

The half wave potential $(E_{0\cdot5})$ of each polarogram was determined and then graph was plotted between $E_{0\cdot5}$ and logarithm of the concentration of the ligand $(\log Cx)$[10]. Linear plot of $E_{0\cdot5}$ *vs.* log Cx (Fig. 1) indicates that a single complex predominates over the entire range investigated[11]. Hence the formula of the complex ion and the dissociation constant K_c can be determined[12] from the following equation

$$(E_{0\cdot5})_c - (E_{0\cdot5})_s = \frac{0\cdot05915}{n} \times \left[\log K_c - \log \frac{f_s k_c}{f_c k_s} - p \log C_x f_x\right] \quad (1)$$

at 25° C

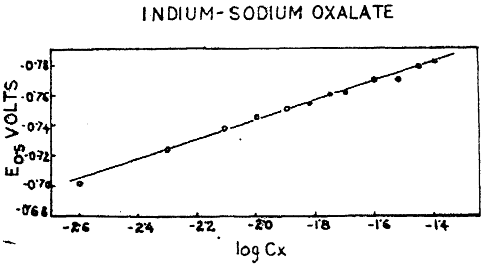

FIG. 1

TABLE I

Change in the half wave potential of indium (III) due to change in the concentration of ligand from 0·0025 *to* 0·0400 *M.*

Sl. No.	Concentration of ligand (M)	Log_{10} C.	$E_{0\cdot5}$ volts	$dE_{0\cdot5}$ volts.
1.	0·0000	0·0000	−0·615	..
2.	0·0025	−2·6021	−0·705	0·090
3.	0·0050	−2·3010	−0·725	0·110
4.	0·0075	−2·1249	−0·738	0·123
5.	0·0100	−2·0000	−0·745	0·130
6.	0·0125	−1·9031	−0·750	0·135
7.	0·0150	−1·8239	−0·753	0·138
8.	0·0175	−1·7570	−0·760	0·145
9.	0·0200	−1·6990	−0·760	0·145
10.	0·0250	−1·6021	−0·770	0·155
11.	0·0300	−1·5259	−0·770	0·155
12.	0·0350	−1·4559	−0·780	0·165
13.	0·0400	−1·3979	−0·783	0·168

It is tacitly assumed[13, 14] that K_c is much smaller than $C^p x$, so that the concentrations of the simple ion and any possible lower complex can be neglected in the presence of excess of the ligand. Differentiation of the equation (1) gives

$$\frac{d(E_{0\cdot5})_c}{d(\log C_x)} = -\frac{0\cdot05915}{n} p \quad (2)$$

provided that activity coefficient f_s, f_c, and f_x and the liquid-junction potential remain constant as C_x is varied and the ratio k_c/k_s also remains constant.

Since the supporting electrolyte strength was constant throughout, other activity coefficients change only very slightly with the change in the ligand concentration. Hence in calculating the dissociation constant (K_e) by equation 1 the activity coefficients were neglected[15].

The value of p calculated by equation 2 for graph 1 is 3·12 indicating the formation of 1:3 indium (III) oxalate complex with dissociation constant $1\cdot905 \times 10^{-13}$.

The outer 'd' shell of the indium ion is complete hence for co-ordination number 6 indium prefers d^2sp^3 hybridization, the six covalent bonds directed to the apices of a regular octahedron (Fig. 2).

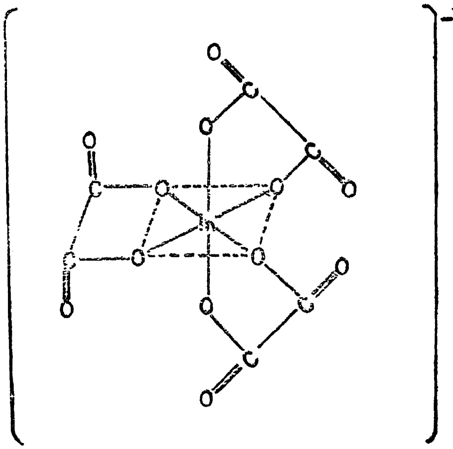

FIG. 2

Thanks are due to Prof. S. S. Nigam, Head of the Department of Chemistry, for giving the facilities.

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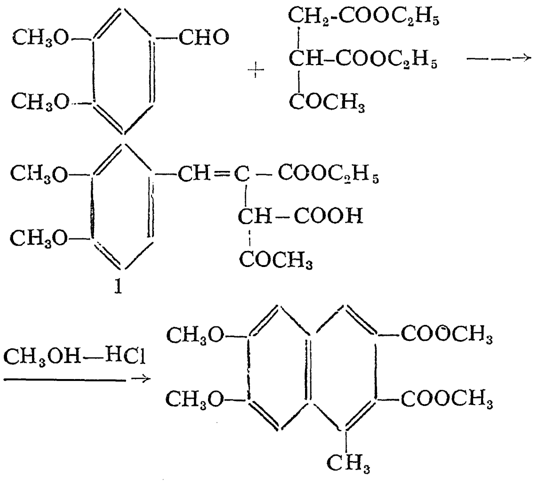
1. Master, D. L., *J. Phys. Chem.*, 1962, **66**, 249.
2. Dutta, N. K. and Bose, P., *J. Ind. Chem. Soc.*, 1953, **30**, 431.
3. Kovalenko, P. N., *Technol. Pokrytii Metallov Metody Kontrolya Proizv* (Rostov-on-Don; Rostovsk Univ.) Sb. 1962, p. 62.

4. Neogi, P. and Dutta, N. K., *J. Ind. Chem. Soc.*, 1938, **15**, 83.
5. Deichman, E. N., *Zhur. Neorg. Khim.*, 1959, **4**, 2360.
6. Dubovenko, L. I., *Ukr. Khim. Zh.*, 1952, **28**, 682.
7. White, J. M., Tang, P. and Li, N. C., *J. Inorg. and Nuclear Chem.*, 1960, **14**, 255.
8. Meites, L., *Polarographic Techniques*, Intersc. Pub. Sec. Ed. 1967, p. 219.
9. Vogel, A. I., *Quantitative Inorganic Analysis*, ELBS, ed., 1963, p. 1009.
10. Meites, L., *Polarographic Techniques*, Inter-Sci. Pub. Sec. Ed. 1967, p. 269.
11. —, *Ibid.*, p. 273.
12. —, *Ibid.*, p. 268.
13. Deford, D. D. and Andersen, D. L., *J. Am. Chem. Soc.*, 1950, **72**, 3918.
14. Buck, R. P., *J. Electro Anal. Chem.*, 1963, **5**, 295.
15. Meites, L., *Polarographic Techniques*, Inter-Sci. Pub. Sec. Ed., 1967, p. 272.

SYNTHESIS OF 1-METHYL-2, 3-DICARBOMETHOXY-6, 7-DIMETHOXYNAPHTHALENE

In a new synthetic approach to the 1-phenylnaphthalene systems of lignans, a model compound, 1-methyl-2, 3-dicarbomethoxy-6, 7-dimethoxynaphthalene has been prepared.



The condensation of veratraldehyde and diethyl acetosuccinate[1,2] in the presence of sodium ethoxide in ethanol gave a mixture of esters in the neutral fraction of the reaction product. However, there was an acid fraction (48%), which gave a crystalline meterial (53·3 %), m.p. 155°. The analytical data (Found: eq. wt. 339; C, 60·42; H, 5·85 %) indicated it to be an acid $C_{17}H_{20}O_7$ (Calc.: eq. wt. 336; C, 60·71; H, 5·95 %); to which the structure (1) was assigned as a compound formed *via* a γ-lactonic route of a Stobbe reaction[3]. Saponification yielded 3, 4-dimethoxyphenyl itaconic acid [3, 4-$(CH_3O)_2$-C_6H_3-CH=C·(COOH)·CH_2·COOH] m.p. 172°; m.m.p. 171· undepressed with an authentic sample[4] m.p. 173–5°; apparently the acetyl group is lost in the saponification.

Treatment with methanolic hydrogen chloride of the acid-ester (1) resulted not only in esterification but also a *trans*-esterification and further cyclization, for it gave a neutral compound m.p. 170°; uv: $\lambda_{\text{max}}^{\text{EtOH}}$ 234 nm (log ε 3·89), 281 nm (log ε 3·57); NMR spectra clearly indicated 2 methyl esters (3H, s, 3·46; 3H, s, 3·60 δ), an arylmethyl (3H, s, 1·83 δ), apart from the aromatic protons (3H, 6·70, 6·87, 7·32 δ) and the aryl dimethoxy groups (6 H, s, 3·87 δ) leading to the structural assignment of the naphthalene system (2) to the compound.

Similarly piperonal yielded 3, 4-methylenedioxy-1-methyl-2, 3-dicarbomethoxynaphthalene m.p. 205–7°; uv: $\lambda_{\text{max}}^{\text{EtOH}}$ 240 nm (log ε 3·70), 288 nm (log ε 3·69); NMR: 2 methyl esters (3H, s, 3·53; 3H, s, 3·62 δ), an arylmethyl (3H, s, 1·92 δ), aromatic protons (3H, 6·70, 6·74, 7·33 δ) and arylmethylenedioxy protons (2H, s, 5·96 δ).

Employment of this reaction sequence to the aroylsuccinates is in progress.

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1. Adkins, H., Isbell, N. and Wojcik, B., *Organic Synthesis*, John Wiley and Sons, Inc., New York, Vol. 2, p. 262.
2. Banerjee, S. K., *J. Indian Chem. Soc.*, 1931, **8**, 779.
3. Johnson, W. S., McCloskey, A. L. and Dunnigan, D. A., *J. Am. Chem. Soc.*, 1950, **72**, 514
4. Campbell, K. N., Cella, J. A. and Campbell, B. K., *Ibid.*, 1953, **75**, 4681.

VANADAMETRIC DETERMINATION OF SULPHANILAMIDE

So far no oxidametric method was used of for the assay of sulphadrugs. Since sodium vanadate is known to be a clean oxidametric reagent for the oxidation of organic compounds, the present authors tried to make use of it for the titrimetric determination of sulphanilamide. Estimation by direct titration has not been found possible in view of the slow reaction between sulphanilamide and sodium vanadate. However, the estimation can be successfully carried out by adding an excess of sodium vanadate to sulphanilamide and back titrating the excess with Fe (II) solution.

Under the experimental conditions, the oxidation of sulphanilamide by the sodium vanadate proceeds according to the equation,

$$(H_2NO_2S)\ C_6H_4\ (NH_2) + 6\ NaVO_3 + 18\ HCl \longrightarrow$$
$$(H_2NO_2S)\ C_6H_4\ (NO_2) + 6\ VoCl_2 + 6\ NaCl + 10\ H_2O$$

To an aliquot volume of sulphanilamide (containing 22–30 mg. of sulphanilamide), 25 ml bf concentrated hydrochloric acid, 5 ml of orthophosphoric acid and 10 ml of sodium vanadate (0·2 M) are added and the volume made upto 50 ml. The solution is kept at room temperature for 45 minutes and then titrated with standard solution of ferrous ammonium sulphate solution using N-phenyl anthranilic acid as indicator. The results obtained (after applying suitable blank correction) are found to have an accuracy better than 0·5%.

Acetanilide which is the starting material for the preparation of sulphanilamide does not interfere in this procedure. Acetate, perchlorate and sulphate ions also do not interfere. Further work regarding the determination of other sulphadrus is in progress.

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METAL CHELATES OF Cu^{2+}, Ni^{2+}, Co^{2+} AND VO^{2+}. A SPECTROPHOTOMETRIC STUDY USING SALICYLALDEHYDE-4-*p*-METHOXY PHENYL-3-THIOSEMICARBAZONE AS A LIGAND

THE communication reports that salicylaldehyde-4-*p*-methoxy-phenyl-3-thiosemicarbazone (SMPT) forms coloured chelates with Cu^{2+}, Ni^{2+}, Co^{2+} and VO^{2+}. Cu^{2+}, Ni^{2+} and VO^{2+} form a 1 : 1 (metal; ligand) chelate while Co^{2+} forms 1 : 2 chelate with this reagent. Effect of pH on chelate and stability constants of chelates has been investigated.

EXPERIMENTAL

The ligand salicylaldehyde-4-*p*-methoxy-phenyl-3-thio semicarbazone was synthesized and crystallised to get an analytically pure sample (m.p. 210)[1]. All the chemicals except the ligand were of BDH analyticals grade.

The solution of ligand was prepared by dissolving required amount of ligand in absolute alcohol. The solutions of metal ions were prepared by dissolving their nitrates (BDH AnalaR grade) except for VO^{2+} for which $VOSO_4$ was used. The concentration of metal ions were checked by standard gravimetric and volumetric methods. All the measurements were carried out on Bosch and Lomb " Spectronic 20" spectrophotometer. pH of the solutions were checked by Phillips pH mater.

RESULTS AND DISCUSSIONS

Effect of pH on the formation of the chelates.—The chelates of the metal ions studied with SMPT are formed within a limited range of pH. The results are included in Table I.

TABLE I

Effect of pH on the formation of the chelates

Chelate	pH range of stability	λ_{max} of chelate (nm)	Studied at pH
Cu (II) SMPT	4·0–7·0	390	4·33
Ni (II) SMPT	4·5–8·2	420	6·99
Co (II) SMPT	5·0–8·0	395	6·12
VO (II) SMPT	4·0–6·0	395	5·33

Nature of the complexes formed.—Mixtures containing metal and ligand in the ratio of 1 : 1, 1 : 2 and 1 : 3 were prepared in 50% (v/v) absolute alcohol. The absorbance measurements were taken from 370 nm. to 600nm, All showed only one λ_{max} as shown in Table I.

Composition of the chelates.—The composition of the metal chelates have been deterimind spectrophotometrically using the method of continuous variation[2] slope ratio[3] and molar ratio[4] methods. In all the systems studied the composition of the complex has been found 1 : 1 (metal : ligand) except for Co^{2+} which forms 1: 2.

The stability constants have been determined by the method of Dey and Banerjee[5] and the mole ratio method at 30° ± 1°. The results are given in Table II.

TABLE II

Stability constants and composition of metal SMPT chelates

Chelate	Composition	Stability constant molar ratio	Stability constant Dey and Banerjee
Cu (II) SMPT	1:1	$2·0 \times 10^6$	$1·9 \times 10^6$
Ni (II) SMPT	1:1	$2·2 \times 10^5$	$1·6 \times 10^5$
VO (II) SMPT	1:1	$2·0 \times 10^5$	$1·2 \times 10^5$
Co (II) SMPT	1:2	$5·1 \times 10^{11}$	$2·7 \times 10^{11}$

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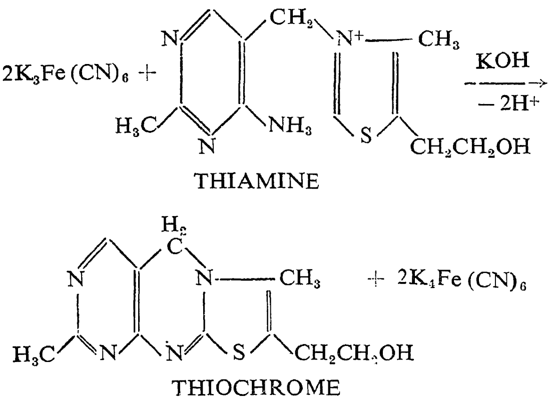
---

1. Shah, I. D. and Trivedi, J. P., *J. Ind. Chem. Soc.*, 1964, **41**, 704.
2. Job, P., *Ann. Chim.* (France), 1928, **9**, 113.
3. Harvey, A. E. (Jr.) and Manning, D. L., *J. Amer. Chem. Soc.*, 1950, **72**, 4488.
4. Yoe, J. H. and Jones, A. L., *Ind. Eng. Chem. Anal. Ed.*, 1946, **16**, 111.
5. Banerjee, S. K. and Dey, A. K., *Z. Analyst. Chem*, 1961, **179** 30.

AMPEROMETRIC DETERMINATION OF THIAMINE (VITAMIN B_1) USING POTASSIUM HEXACYANOFERRATE (III)

Introduction

Thiamine has been estimated[1-3] by polarographic, chromatographic, colorimetric, physico-chemical, enzymatic and microbiological methods. All these methods are either time consuming and tedious and have low accuracy. Thiamine is known to undergo oxidation[4] when treated with alkaline hexacyanoferrate (III) (pH around 12) to give thiochrome. Preliminary studies show that one mole of thiamine is oxidised by two moles of hexacyanoferrate (III) according to the following equation:



On the basis of this reaction, thiamine has been estimated amperometrically using potassium hexacyanoferrate (III). The method was successfully employed for the estimation of thiamine in multivitamin tablets and Elixir Eupeptine.

Experimental

Solutions were prepared using AnalaR grade (BDH) chemicals. Ferricyanide solution was standardised by the potassium iodide method[5].

Each multivitamin tablet had 0·3 mg of thiamine along with other vitamins and minerals and 10 ml of Elixir Eupeptine syrup contained 0·3 mg of thiamine. Amperometric titrations were carried out with a manual Toshniwal (India) polarograph (CLO–2) using a Pye scalamp galvanometer and a dropping mercury electrode (capillary characteristic: 1·5 mg^{2,3} ^{1,2}) and LiCl as supporting electrolyte. The effective concentration of LiCl should be between 1·0 *M* and 1·5 *M*.

Procedure

0·5 ml of thiamine solution (concentration between 0·0113 to 0·000113 *M*) was placed in the polarographic cell. To this was added 5·0 ml of 0–05 *M* KOH and 10 ml of 2·0 *M* LiCl and the solution was deaerated by bubbling purified hydrogen. It was then titrated amperometrically against 0·1 to 0·001 *M* $K_3Fe(CN)_6$ at –0·35 V.

Table 1

Estimation of thiamine by amperometric titration against $K_3Fe(CN)_6$

	Thiamine mg/100 ml Taken	Thiamine mg/100 ml Found	Accuracy %	Precision* %
	300	301·01	0·3	0·6
	30	29·88	0·4	1·0
	20	19·85	0·7	1·1
	10	10·15	1·5	0·9
	5	5·07	1·4	2·2
	3	2·94	2·0	1·6
	2	1·95	2·5	3·0
Thiamine in multivitamin tablets				
Glaxo	2·4	2·36	1·5	2·5
Squibb	2·4	2·35	2·1	2·4
Thiamine in Elixir Eupeptine				
	2·4	2·37	1·2	2·5

* Based on the results of triplicate analysis.

For the estimation of thiamine in multivitamin tablets, the powder of two tablets was shaken up with 10 ml of double distilled water and filtered. The filtrate was diluted to 25 ml and 2·0 ml of this solution was taken in the polarographic cell and to this was added 6·0 ml of 0·05 *M* KOH and 12 ml of 2·0 *M* LiCl. This solution was titrated amperometrically at −0·35 V against 0·001 *M* $K_3Fe(CN)_6$ as above. For the estimation of thiamine in Eupeptine, 10 ml of the syrup was diluted to 25 ml with distilled water. Any precipitate that settled down was filtered off. The filtrate (2·0 ml) was taken in the polarographic cell and titrated amperometrically at −0·35 V against 0·001 *M*-$K_3Fe(CN)_6$ as already described.

Results and Discussion

The amperometric titrations show that 0·02 mg/ml of thiamine can be determined by this method with an accuracy of 2·5% and a precision of 3%. Sharp inflections are obtained even with very dilute solutions. The method has been successfully employed to determine thiamine in multivitamin tablets and in Eupeptine syrup, *i.e.*, in the presence of other vitamins, with a high degree of accuracy (Table I).

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1. Horwitt, M. K., *Ann. Rev. Biochem.*, 1959, **284**, 411.
2. Michelsen, O. and Yamamoto, R. S., *Method Biochem. Anal.*, 1952, **6**, 191.
3. Pleticha, R., *Anal. Chem.*, 1952, **24**, 916.
4. West and Todd, *Text Book of Biochemistry*, Macmillan, New York, 2nd Edition, p. 704.
5. Vogel, A. I., *Text Book of Quantitative Inorganic Analysis*, Longmans Green, London, 1968.

SOME OBSERVATIONS ON REPRODUCTION OF *RHINOLOPHUS ROUXI ROUXI* (MICROCHIROPTERA)

LITTLE information is available on the breeding habits of the tropical members of the family Rhinolophidae. The reproductive behaviour of the European species of this family, *viz.*, *Rhinolophus ferrum equinum*[1] and *Rhinolophus hipposideros*[2] is like that of other temperate bats. They usually mate in fall and spermatozoa are stored in the uterus and vagina through the winter. Ovulation and fertilization occur in the following spring.

This family is represented in India by the Rufous Horse-shoe bat, *Rhinolophus rouxi rouxi* (Temminck). Very little is known about the reproductive behaviour of this species except for casual records of pregnancy[3]. The foetal membranes of *Rhinolophus rouxi rouxi* are described by Gopalkrishna[4].

The present note records some observations on reproduction in *Rhinolophus rouxi rouxi.* The material consists of a number of collections of both sexes made through the year. They were collected from their wild state, about 40 miles from Bangalore.

Observations

In the collections made, 63% happen to be females; there thus appears a disproportionate sex ratio in favour of this sex. *Rhinolophus rouxi rouxi* experiences only one well defined breeding season a year. A study of the tooth wear reveals that sexual maturity is reached a year after birth.

Sperms are found in the testes weighing 20–25 mg. Bats with their testes in this weight range and above are functionally active during the breeding season. The testes begin to increase in weight in July and reach their maximum weight of 45–50 mg during October/November, and from December onwards gradually decrease in weight. The cauda-epidydymids are gorged with spermatozoa only during the breeding season. The testes and the accessory organs of reproduction undergo regression following cessation of sexual activity; the period of quiescence lasts till July/August following.

The uterus of *Rhinolophus rouxi rouxi* is bicornuate. The right ovary and the right uterine cornu are larger than the left during sexual maturity. Copulation occurs during late November/early December. Only one ovum is released. Copulation is immediately followed by fertilization and development of the embryo. Ovulation is invariably from the right ovary. During this period the copulated females generally contain cleaving eggs or morulae in the fallopian tubes and uteri respectively.

Spermatozoa are not stored in the genital tract of the female bat during any other part of the year.

The corpus luteum is single, large and occupies the major part of the ovary; occasionally it lies outside the confines of the ovary. The ovary is enclosed in a complete ovarian capsule. In each pregnancy there is only one foetus. Gestation takes place in all cases in the right uterine cornu. The young ones are born during late April/early May after a period of about 150 days gestation. Parturition in most cases is by head presentation and in rare cases by breech presentation. The placenta is expelled about half an hour after birth of the young bat.

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---

1. Mathews, L. H., *Trans. Zool. Soc.*, 1937, 23, 213.
2. Baker, I. R. and Zita Baker, *J. Linn. Soc. Zool.*, 1936, 40, 123.
3. Brosset, A., *Mammalia*, 1962, 26, 166.
4. Gopalkrishna, A., *Curr. Sci.*, 1974, 43, 516.

UTILIZATION OF RESPIRATORY SUBSTRATES BY *ANACYSTIS NIDULANS* RICHTER

Some of the blue-green algae such as *Anabaenopsis circularis*, *Calothrix grevisima*, *Chlorogloea fritschii*, *Nostoc muscorum*, *Phormidium luridium*, *Plectonema calothricoides* and *Tolypothrix tenuis* have been reported to grow well in the dark[1]. But, some like *Anacystis nidulans* and *Anabaena variabilis* are obligate photo-autotrophs and they do not show any response in the dark to externally added carbon sources[2]. In the present study we report the response of the dark starved cells of *A. nidulans* to common respiratory substrates.

Table I

Utilization of some common respiratory substrates by dark starved cells of Anacystis nidulans

Respiratory substrates[1]	lag period	μ moles of O_2 consumed/hr/3×10^7 cells
Control[2] (Endogenous rate)	nil	546
Glucose	60 min.	420
α-ketoglutarate	2 min.	504
Succinate	2 min.	588
Pyruvate	2 min.	756

[1] 50 μm of the appropriate substrate were incorporated into the reaction mixture.

[2] When 50 μm $NaHCO_3$ was used 3×10^7 cells liberated 1146 n moles of oxygen/hour.

Pure culture of *A. nidulans* (Nr. 14011, IARI, New Delhi) was grown at 30° C under 12 hr illumination (5,000 lux) and 12 hr dark period. Cells from a 3-day old shake cultures were taken for study. The cells were taken at the end of 12 hr illumination and starved in the dark for 6 hours before the addition of respiratory substrates. Oxygen exchange by these cells was monitored at 30° C using a Beckman oxygen analyser connected to a Heath Servo Recorder (Model EU-20 B). Oxygen evolution and consumption were measured in 3 ml medium containing 100 mM phosphate (pH 7·0), 10 mM $MgCl_2$ and 50 μM of the appropriate substrate. The instrument was calibrated with water saturated with air at 30° C with full scale deflection (237 μM O_2).

The results are presented in Table I. The response of cells was similar to the endogenous rate of O_2 consumption when succinate and α-ketoglutarate were used as substrates. When glucose was used as the respiratory substrate the response was markedly lower. Also the cells required 60 minutes to show any response to the added glucose. A marked increase in the O_2 consumption could be noticed when pyruvate was used as the respiratory substrate. These results indicate that *A. nidulans* can be grown heterotrophically in the dark using suitable substrates such as pyruvate and it can no more be considered as an obligate photoautotroph.

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---

1. Khoja, T. and Whitton, B. A., *Arch. Mikrobiol.*, 1971, 79, 280.
2. Carr, N. G., *The Biology of Blue-green Algae*, Blackwell Scientific Publications, U.K., 1973, p. 62,

MUTATION FREQUENCY IN RELATION TO M_1 STERILITY

It is well known the mutagen treated material show varying degrees of sterility in M_1 generation[1-4]. An attempt was made to see whether the M_1 plants showing comparable degrees of sterility under the influence of ethyl methane-sulfonate (EMS) and X-rays, singly or in combination, yield proportionate amount of mutations.

Dry seeds of black gram (*Phaseolus mungo* L.) and cowpea (*Vigna sinensis* L. Savi) were treated with X-rays and EMS singly as well as in combination as described elsewhere[5]. The M_1 plants were grouped into five sterility groups (Table I) and the mutation rate was determined separately from each group. The M_1 plants obtained after EMS treatment mostly fell under group A, while those obtained after X-irradiation could be grouped under A and B groups, following combined treatments most of the M_1 plants were in A, B and C groups.

TABLE I

Frequency distribution of M_1 plants under different sterility groups (in percentage)

Mutagenic treatment	Sterility groups (in percentage)				
	A 0–20	B 21–40	C 41–60	D 61–80	E 81–100
Black gram					
X-rays	55·7	20·5	10·7	8·3	4·8
EMS	66·6	19·5	7·0	4·4	2·2
X-rays + EMS	33·2	38·0	19·2	12·8	6·8
Cowpea					
X-rays	42·2	22·2	17·8	12·2	5·6
EMS	56·7	22·8	11·4	5·7	3·4
X-rays + EMS	27·7	28·6	20·6	15·5	7·6

TABLE II

Chlorophyll mutation frequency in relation to sterility in black gram and cowpea

Mutagenic treatment	Mutation frequency				
	A 0–20	B 21–40	C 41–60	D 61–80	E 81–100
Black gram					
X-rays	0·26	1·56	2·44	0·61	0·10
EMS	2·86	1·98	0·43	0·12	0·03
X-rays + EMS	1·64	3·83	2·88	1·20	0·18
Cowpea					
X-rays	0·32	1·68	2·36	0·43	0·10
EMS	2·58	1·83	0·54	0·20	0·04
X-rays + EMS	1·78	3·62	3·10	1·68	0·26

The mutation frequency in different sterility groups depended on the nature of mutagen (Table II). In X-irradiated progeny, the mutation rate was very high in B and C groups and was comparatively low in the other sterility groups. After EMS treatment, the mutation rate was very high in A and B, *i.e.*, low sterility groups, as the sterility increased, the mutation rate showed gradual decrease. With combined treatments, the mutations were found in all the groups, the highest being in B and C groups and a decline in A and D groups. For X-rays, there was an obvious 'threshold value' *i.e.*, no mutations appear without a noticeable decrease in M_1 fertility, possibly owing to the fact that the X-ray induced sterility rose more or less linearly with dosage, as do mutations. With EMS, there was a definite 'threshold value', *i.e.*, mutations occur in the offspring of M_1 plants without any decrease in fertility. Combined treatments gave a picture like the one obtained after EMS. Definitely higher threshold values seem to be obtained with EMS or with combined treatments than with X-rays.

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1. Ehrenberg, L., Gustafsson, A. and Lundqvist, U., *Hereditas*, 1961, **47**, 243.
2. Ekerberg, I., *Ibid.*, 1969, **63**, 257.
3. Gaul, H., *Rad. Bot.*, 1964, **4**, 155.
4. Gilot-Delhalle, J. and Moutschen, R., *Ibid.*, 1972, **12**, 381.
5. Jana, M. K. and Appa Rao, S., *Indian J. Genet*, 1974, **34**, 32.

ISOLATION OF INGENOL FROM THE IRRITANT LATEX OF *EUPHORBIA SERRATA* L.

LIKE other plants of the Spurge family, the plants of *Euphorbia serrata* exude a milky, sticky, caustic and skin irritant latex when the leaves or the stems are cut or broken[1]. From the latex of this plant, which has pronounced cocarcinogenic activity[2], we wish to report the presence of ingenol, the fatty acid esters of which (and of its 16-hydroxy derivative) are known to have cocarcinogenic activity on mice skin[3,4].

The methanolic latex preparation, following the standard method[5], gave an irritant mass; ID$_{50}$: 1·5 μg/ear[6], which was defatted and hydrolysed with 0·5 M KOH to give the parent diterpene ingenol[5]. Its identity was confirmed by converting it to ingenol-3, 5, 20-triacetate[5] : $C_{26}H_{34}O_8$ (MS), parent ion m/e 474, UV (MeOH) : λ_{max}, 212, 290 ; $\epsilon = 16300$, 220 ; IR (CH_2Cl_2) : 1740, 1505, 1640 cm^{-1}, relative retention time on 5% QF$_1$ column (GLC) : 3·8 min[7]. These data are in agreement with those of an authentic sample of ingenol triacetate.

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Iran, *October* 25, 1975.

---

1. Morton. J. I., *Plants Poisonous to People.*, Hurricane Pub. Inc., Miami, Florida. 1971.
2. Upadhyay, Rajiv R, and Zarintan, M. H., *Experientia* (Tobe published).
3. — and Hecker, Erich, *Phytochemistry*; 1975, 24, 2514.
4. Adolf, W. and Hecker, E., *Z. Krebsforsch.*, 1975, 84, 325.
5. Upadhyay, Rajiv R., Zarintan, M. H. and Ansarin, M., *Planta Medica* (In press).
6. Hecker, E., In : *Methods in Cancer Research*, Ed. H. Bush, Academic Press, N.Y., 1971, 6, 438.
7. Upadhyay, R. R., *Chromatographia* (In press).

BREAKING DORMANCY IN POTATOES BY VAPOUR TREATMENT WITH ETHYL ALCOHOL

CHEMICAL treatments to break the dormancy in seed potatoes can be effected either through soaking in aqueous solutions or by exposure to vapours of volatile chemicals. Thiourea, ethylene chlorohydrin and gibberellic acid, singly or in combination, are field proven chemicals for the former method, while ethylene chlorohydrin and carbon disulphide are for the latter[1-4].

Ethylene chlorohydrin, which is preferentially used at this Institute for vapour treatment of dormant seed tubers[5], has pronounced human toxicity; it is also expensive and is not indigenously produced. In the course of a search for alternate chemicals, we found the vapours of the low homologues of aliphatic alcohols to by effective and free from the above defects.

A pilot project study of the efficacy of vapour treatment with ethyl alcohol for breaking seed-tuber dormancy was undertaken in the autumn of 1974 at Kufri Station. Five quintals each, of the freshly harvested seed of ‘Kufri Muthu’ variety were used in the vapour treatment with ethyl alcohol (95%) and compared with ethylene chlorohydrin; both were used at the rate of 2 ml per kg of tubers in an air-tight chamber[4]. The chemicals were kept in shallow dishes, above which gunny bags containing the seed were stacked and at frequent intervals the air within the chamber was circulated with a fan. The treatment lasted for 48 hours; the temperature within the chamber was maintained around 25° C for the entire duration of the treatment.

The treated tubers were planted at Daurala on 25–9–1974 in randomised blocks of 8 replications with the above two treatments along with an untreated control in plots measuring 7 m by 4·2 m. Periodic plant emergence counts were taken commencing from 45 days after planting. Table I gives the results.

It could be seen that vapour treatment with ethyl alcohol was as effective as the ethylene chlorohydrin.

TABLE I

Progressive emergence, final stand, Bartlett's rate index and tuber yield for the vapour treatments

	Progressive % emergence, days after sowing					Final stand (%)	Bartlett's rate index	Yield of tubers (q/ha)
	45	52	59	66	73			
Vapour treatment with ethylene chlorohydrin	1·3	23·3	47·4	49·6	81·7	93·0	0·5312	183
Vapour treatment with ethyl alcohol	0·4	24·9	47·4	52·9	81·3	94·6	0·5412	215
Control	..	1·8	1·3	35·0	46·9	50·2	0·4288	91
S.E.								±19
C.D. at 5% level								58

With our present findings of the effectiveness of ethyl alcohol for breaking the dormancy in buds, a clearer picture of the " structure-activity " relationship among at least the three chemicals, ethyl alcohol, ethylene chlorohydrin and " Ethrel" (2-chloroethane phosphonic acid) all of which possess, in common, the C_2-frgament CH_2–CH_2-emerges. It is our conjucture that this C_2-fragment gives rise, *in vivo*, to ethylene (CH_2–CH_2) which has been established to be an endogenous growth hormone.[7] Further evidence of the effect of ethylene on potato dormancy and sprout growth is available from the resent work of Rylaski *et al.* (1974)[8].

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October 28, 1975.

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1. Saptharishi, K. and Azariah, M. D., *Madras agric. J.*, 1953, **40**, 414.
2. Ramanujam, S., Singh, M. and Swaminathan, K., *Indian J. agric. Sci.*, 1957, **27**, 35.
3. Azariah, M. D. and Rai, R. P., *Indian Potato J.*, 1960, **2**, 100.
4. Purohit, A. N., *Indian J. Physiol.*, 1970, **13**, 185.
5. *Scientific Report of the Central Potato Research Institute for the Years* 1961 *and* 1962. New Delhi, Government of India, Ministry of Food and Agriculture.
6. *Ibid.* for the year 1963.
7. Pratt, H. K. and Goeschl, J. S., *Ann. Rev. Plant Physiol.*, 1969, **20**, 541.
8. Rylski, I., Rappaport, L. and Pratt, H. K., *Plant Phasiol.*, 1974, **53**, 658.

EMS AND MMS INDUCED LEAF MUTATIONS IN PEAS

MANY workers have reported the occurrence of leaf mutations in mutagenic experiments and there is a fair indication that these are true mutations[1, 2, 12]. In the present studies a few types of leaf mutants are reported.

Pea seeds of Bounvile (*Pisum sativum* L.) and two mutagens, ethyl methane sulphonate (EMS) and methyl methane sulphonate (MMS) were used during these studies. From the preliminary studies, only one dose of the two mutagens, *viz.*, EMS 0·25% and MMS 0·025% were tried[9]. Pea seeds were treated with mutagenic solutions for 10 hours and sown. One hundred seeds were treated during M_1 and there were 2510 plants in M_3 generation. The observations recorded during M_1, M_2 and M_3 are expressed as percentage of total populations.

The data (Table I) indicate the presence of leaf mutations in both EMS and MMS treatments. These mutants comprised of small and narrow leaflets, and leaflets with acute and obtuse apex. Further, it was apparent that the change was more frequent from leaflet condition to simple leaf state. Similar leaf mutations have also been reported by other workers. Fetner[1] observed an enlargement in length and breadth of leaflets in gamma-irradiated *Graptopetalum macdougallis*. An increased number of leaflets have been reported by Stubbe[12] in irradiated soybean and *Vicia.* Narrow leaflet mutants with X-ray have been observed by Gottschalk[2] which proved to be high yielders. Jones[6] obtained dwarf, crinckled leaves and exart long peduncle mutants in *Vigna sinensis.* The percentage of leaf mutants in M_1, M_2 and M_3 varied from 0·33% to 2·7% in EMS and from 0·46% to 3·0 in MMS treatments. The distribution of different types of mutants appeared to be at random. These leaf

TABLE I

Leaf mutations in EMS and MMS treated peas

Treatment	Generation	Unequal leaflets %	Small leaflets %	Leaflets with acute apex %	Leaflets with obtuse apex %	Total %
EMS 0·25%	M_1	1·8	0·6	0·6	0·3	3·3
	M_2	0·3	nil	0·9	0·3	1·5
	M_3	nil	nil	1·5	1·2	2·7
MMS 0·025%	M_1	0·18	0·17	0·11	nil	0·46
	M_2	2·4	0·3	nil	0·3	3·0
	M_3	0·6	0·4	0·4	nil	1·4

mutations segregated in M_2 and M_3 generations indicating that these were due to genic changes; but due to low plant populations no definite ratio could be obtained. However, a few investigators have reported the breeding behaviour in other plants. Segregation of leaf mutations in *Vicia faba* into unifoliata and normal types have been reported by Sjodin[11]. Patil and Bora[10] noted that small and narrow leaf mutants in groundnut were recessive in nature. Hammons[4] and Loesch[7] observed a 3:1 ratio in groundnut leaflet mutants. Besides, the genic segregation, cytological evidence indicates that the leaf mutations arise due to alterations in chromosome pattern as has been reported by Husted[5], Mendes[8] and Gustaffsson and Gadd[3].

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1. Fetner, R. H., *Radiation Research*, 1965, **5**, 365
2. Gottschalk, W., *Radiation Botary* (*Suppl.*), 1965· **5**, 385.
3. Gustaffsson, A. and Gadd, I. *Hereditas*, 1967, **53**, 143.
4. Hammons, O. R., *Crop Science*, 1964 **4**, 22.
5. Husted, L. *Cytologia*, 1936 **7**, 396.
6. Jones, S. T., *J. Heredity*, 1965, **56**, 273.
7. Loesch, P. J. Jr., *Crop Science*, 1964, **4**, 73.
8. Mendes, A. J. I., *Bragantia*, 1947, **7**, 257.
9. Narsinghani, V. G. and Sudhir Kumar, *Theoret. and Appl. Genet.*, 1971, **41**, 18.
10. Bora, S. H. and Bora, K. C., *Indian J. Genet.*, 1963, **23**, 47.
11. Sjodin J., *Hereditas*, 1971, **67**, 155.
12. Stubbe, H., *Indian J. Genet.*, 1959, **22**, 192.

CHANGES IN THE FREE AMINO ACIDS OF CITRUS LEAVES IN RELATION TO CITRUS GREENING AND CITRUS CANKER

WITH a view to assessing the quantitative and qualitative changes in the amino acid composition of the diseased and the healthy plants, two commonly occurring diseases of commercially grown citruses were studied; canker disease on kinnow and greening disease of musambi, were included in this study and the results are presented in this paper.

Healthy and diseased leaves of the same maturity were used for analyses. The method of extraction and determination of amino acids were the same as described by Rangaswami and Natrajan[1]. The data from the analyses are presented in Table I.

TABLE I

Amounts of amino acids (in mg/g fresh leaves) in healthy and diseased leaves of kinnow and musambi

Amino acids	Kinnow (canker)		Musambi (greening)	
	Healthy	Diseased	Healthy	Diseased
Glutamic acid	1·1212	trace	0·728	0·627
Aspartic acid	0·1714	0·805	0·182	0·203
Serine	0·143	0·009	trace	0·084
Glycine	0·857	0·537	0·91	0·677
Asparagine	trace	trace	trace	trace
Proline	0·857	0·268	0·227	2·091
Methionine	0·143	0·089	trace	trace
Valine	1·1212	0·537	0·7705	0·338
Phenylalanine	0·428	0·805	0·546	0·340
Unknown	..	trace	..	..

The number of amino acids in both the diseased and healthy leaves is the same except in case of canker on kinnow an unknown amino acid was located. In general, the quantities of amino acids in the diseased leaves were lower than those of the healthy leaves. Increase in the quantities of amino acids in the canker affected kinnow leaves with corresponding decrease in musambi affected by greening was noticed in aspartic acid and phenylalanine. Contrary to this an increase in quantity of an amino acid in the disease over healthy in greening with corresponding decrease over control in canker was noted in serine and proline.

Thus the amino acid content of the leaves cannot be correlated with the disease in these plants.

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December 26, 1975.

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1. Rangaswami, G. and Natrajan, S., *Indian Phytopath.*, 1966, 19, 59.

REVIEWS AND NOTICES OF BOOKS

Methane : Proceedings of a One-day Seminar. Compiled by Dr. Leo Pyle and Peter Fraenkel. (Intermediate Technology Publications Ltd., 9 King Street, London, WC2E 8 HN, U.K.), 1975. Pp. 51. Price not mentioned.

The recent escalation in the price of oil has led to a global energy crisis and has had a shattering impact on the economies of most of the developing countries. There has been the sudden realisation all over the world that the era of unlimited supplies of cheap energy is over. Therefore there is a great urgency for tapping alternate sources of energy and it is in this context that the Intermediate Technology Development Group (ITDG) in U.K. is doing commendable work. This group consisting of engineers, economists, scientists and others from industry and the professions is interested in the development of effective self-help techniques and low cost technologies using local resources of the poor countries of the world. The group has set up a power panel to study the possible contribution to the energy supply of methane generation produced by the anaerobic fermentation of animal and vegetable wastes. In this small booklet under review are summaries of the proceedings of a one-day discussion meeting organised by this group. The range of papers discussed cover the development, manufacture and marketing of anaerobic fermenters. The summaries make interesting reading giving rise to the expectation that the day may not be far off when methane generation would play a vital role not only as a source of energy and fertiliser but also as part of the waste disposal cycle of rural communities. It is to be hoped that further developments in this field would find quick publication and be made available to all those interested in them.

K. M. Sivanandaiah.

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In view of the Labour Strike in The Bangalore Press we are sorry we could not bring out the Journal for July 5, 1976 in time. The inconvenience caused is very much regretted.

Bangalore 56006
July 15, 1976

Manager
Current Science

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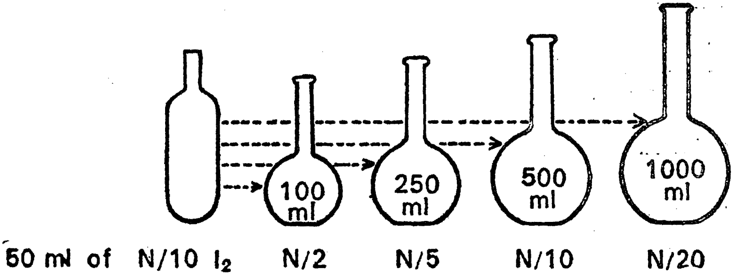

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COLORIMETRIC STUDIES ON THE REACTIONS OF DIOXOURANIUM (VI) WITH 2, 4-DIHYDROXYVALEROPHENONE AND 2, 4-DIHYDROXYCAPROPHENONE

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ABSTRACT

Dioxo-uranium (VI) forms orange yellow complexes with 2, 4-dihydroxyvalerophenone and 2, 4-dihydroxycaprophenone having λ_{max} at 400 nm in the pH range 4·0–4·5 and 4·3–4·8 respectively. The composition of the complexes as 1 : 2 (metal : ligand) has been determined by continuous variation, mole ratio and the slope ratio methods. The molecular extinction coefficients of the complexes are $3{\cdot}20 \times 10^2$ and $2{\cdot}95 \times 10^2$ at 400 nm and the stability constants are $3{\cdot}981 \times 10^5$ and $3{\cdot}622 \times 10^5$ with 2, 4-dihydroxyvalerophenone and 2, 4-dihydroxycaprophenone respectively. The limit of interference due to the presence of foreign ions in the colorimetric determination of uranium has also been determined.

INTRODUCTION

A NUMBER of reagents[1–5] have been used for colorimetric studies of uranium. This communication deals with the colorimetric studies on the reactions of 2, 4-dihydroxyvalerophenone (DHV) and 2, 4-dihydroxycaprophenone (DHC) with dioxo-uranium (VI).

EXPERIMENTAL

Reagenis :

DHV[6–8] and DHC were synthesized by condensing *n*-valeric/*n*-caproic acid with resorcinol in presence of anhydrous zinc chloride at 160° C and crystallised from a mixture of hexane and toluene. They were characterized by their NMR spectra in $CDCl_3$. The deformed triplet at $9{\cdot}10\ \tau$ with $J = 6{\cdot}0$ Hz was assigned to methyl group of the alkyl chain and the methylene protons α to $C = O$ appeared as triplet at $7{\cdot}14\ \tau$; $J = 7{\cdot}5$ Hz, while the rest of the methylenes appeared as multiplet at $8{\cdot}0$–$9{\cdot}10\ \tau$. The multiplet at $3{\cdot}50$–$3{\cdot}75\ \tau$ stands for two aromatic protons *H*-3 and *H*-5 and a doublet at $2{\cdot}40\ \tau$ with $J = 9{\cdot}0$ Hz for *H*-6 proton. Two D_2O exchangeable hydroxy groups appeared as multiplet at $2{\cdot}90 - 3{\cdot}7\ \tau$ in DHC. DHV also had the same pattern of peaks except that one methylene group was less.

I.R. spectra showed strong bands at 3500 cm^{-1} (OH), 3380 cm^{-1} (chelated OH), 2950 cm^{-1}, 2850 cm^{-1} and 2800 cm^{-1} (methylenes), 1675 cm^{-1} ($C = O$) and 850 cm^{-1} (trisubstituted benzene). A solution of the reagents in 50% ethanol was used in all experiments.

Stock soluton of uranium was prepared by dissolving uranyl nitrate (B.D.H., AnalaR) crystals in double distilled water. Ammonium hydroxide and acetic acid were used to prepare the buffers of different pH values. Ionic strength was maintained by adding an appropriate quantity of 1·0 M solution of sodium perchlorate. All other chemicals were of B.D.H. AnalaR grade

OBSERVATIONS AND RESULTS

DHV and DHC instantaneously form orange yellow coloured solube complexes with dioxo-uranium (VI).

Nature of the Complexes:

Vosburg and Cooper's method[9] employed to the orange yellow solution containing 1:1, 1:2, 1:3 and 2:1 mole ratio of metal to DHV/DHC showed that the reagents form only one complex with uranium which exhibit maximum absorbance at 400 nm in the pH ranges 4·0–4·5 and 4·3–4·8 respectively.

Effect of pH and Reagent :

The uranium complexes were found to be stable in the pH ranges 4·0–5·0. Above pH 5·0 the complexes got hydrolysed resulting in the precipitation of uranium hydroxide. The complexes attained maximum colour intensity in the pH range 4·0–4·5 and 4·3–4·8 for DHV and DHC respectively. Subsequent studies have been carried out at pH 4·2 and 4·7 respectively. It was also found that about 20 fold excess of the reagents was necessary to attain the maximum colour intensity.

Composition of the Complexes:

In all the measurements requisite amounts of ethanol were added to maintain the concentration of ethanol at 25% in final dilution; pH was adjusted as mentioned above and the ionic strength was maintained at 0·1 M $NaClO_4$. The composition of the complexes was determined by Job's method[11], mole ratio and slope ratio methods[11,12]. The results from all the methods indicated the formation of 1:2 (metal: ligand) complexes. In mole ratio method the zero slope of the colour intensity was approached with a large excess of the reagent. It was due to the highly dissociating nature of the complexes.

Stability Constants of the Complexes :

The stability constants of the complexes were calculated from the data in molar ratio method, using the relations :

$$K = C(1 - \alpha)/(m\alpha C)^m (n\alpha C)^n \text{ and}$$

$$\alpha = (Em - Es)/Em$$

where m, n, C, α, Em and Es have their usual meanings. The stability constants were found to be $3{\cdot}981 \times 10^5$ and $3{\cdot}622 \times 10^5$ for DHV and DHC complexes respectively. The standard free energy of formation ($\triangle F = - RT \ln K$) were found to be 8·301 and 7·718 Kcal/mole at 30° C respectively.

Molar Extinction Coefficient and Sensitivity:

The values of molar extinction coefficients from the slope ratio curves are $3 \cdot 2 \times 10^2$ and $2 \cdot 95 \times 10^2$ at 400 nm and the sensitivities are 0·743 and 0·801 μg U/cm^2 (Sandell Scale)[13] for DHV and DHC complexes respectively.

Procedure for Estimation:

The solution containing 30–85 ppm of uranium was treated with 20-fold excess of the reagents and the pH was adjusted as mentioned above. Requisite amount of ethanol was added to maintain its proportion at 25% level in final dilution and the ionic strength was maintained at 0·1 M $NaClO_4$. The absorbance was measured at 400 nm against a blank containing corresponding concentration of metal ions. From the abosrbance value, concentration of the metal ions can be evaluated with the help of the calibration curve.

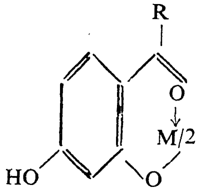
Effect of Foreign Ions:

The influence of foreign ions on the estimation of uranium using DHV and DHC was also studied in the usual manner. It was observed that at 40 ppm concentration of uranium, 1000 ppm of Al (III), Ba (II) Be (II), Cd (II), Ca (II), Fe (II), Mg (II), Sr (II), K (I), Na (I), NH_4 (I), Cl^-, ClO_4^-, NO_3^-, SO_4^{2-}; 750 ppm of Br^-, Co^- (II); 500 ppm of Cr (III), Pb (II), Ag (I), Mn (II), Ni (II); 150 ppm of I^-, Tl (I) could be tolerated. However, Cu (II), Fe (III) and Pd (II) interfere at all levels. These must be eliminated prior to estimation.

Structure of the Complexes:

The structures of the complexes have been proposed on the basis of I.R. spectra. The spectral data for the ligands and the complexes indicate that most of the frequencies are perturbed on complex formation. I.R. spectrum of the ligand shows two bands in OH stretch region assigned to two different types of hydroxyl groups present in the ligand. The band at 3450 cm^{-1} remains unaffected on complexation, hence it may be due to the 4-hydroxy group. The position of the second band (3330 cm^{-1}) and its disappearance in the spectra of the complexes shows that it may be due to the presence of an intramolecularly hydrogen bonded OH at position-2 in the ligand. The co-ordination of the keto group through oxygen is indicated by the lowering of C = O band from 1675 cm^{-1} in the ligand to the range 1625 cm^{-1} in the complexes. The U = O band appears at 920 cm^{-1}.

Appearance of new band at 555 and 560 cm^{-1} is fairly attributed to metal oxygen bands. On the basis of the above evidence, the following structure (I) may be assigned to the complexes:



(I)

M = O = U = O

R = $- C_4H_9$, C_5H_{11}

The results presented above show that DHV and DHC are good reagents for uranium. The method is rapid and reliable and the reagents can be used in acidic medium so that the metal hydroxides are not precipitated. Despite the inductive effect of alkyl chain attached to the carbonyl carbon and hydroxyl group at position-4 in the ligand which increase the stability of the complexes by increasing the basic strength of the ligands, the overall steric effect of the bulkier group (solvation effect) decreases the stability of the DHC complex.

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1. Gupta, B. D. and Malik, W. U., *J. Ind. Chem. Soc.*, 1970, **47** (2), 145.
2. Desai, M. N. and Desai, B. M., *Curr. Sci.*, 1965, **34**, 244.
3. Poddar, S. N., *Ind. J. Appl. Chem.*, 1964, **27**, 67.
4. Urs, M. K. and Neelkantam, K., *J. Sci. Ind. Res.*, 1952, **11 B**, 79.
5. Singh, J. and Gupta, S. P., *Curr. Sci.* (in press).
6. Nogami, H., *J. Pharm. Soc. Japan*, 1941, **61**, 46.
7. Singh, J., Gupta, S. P. and Malik, O. P., *Ind. J. Chem.*, 1975, **13**, 1217.
8. Dohme, A. R. L., Cox, E. H. and Ellis Miller, *J. Am. Chem. Soc.*, 1926, **43**, 1688.
9. Vosburg, W. C. and Cooper, G. P., *Ibid* 1941, **63**, 437.
10. Job, P., *Ann. Chim.*, 1928, **9**, 113.
11. Yoe J. H. and Jones, A. L., *Ind. Eng. Chem. Anal. Ed.*, 1944, **16**, 111.
12. Harvey, A. E. and Manning, D. L., *J. Am. Chem. Soc.*, 1950, **72**, 4488.
13. Sandell, E. B., *Colorimetric Determination of Traces of Metals* (Interscience Publications, Inc., New York), 1959, pp. 83, 97.

THE FISCHER INDOLE SYNTHESIS: 4-NITRO-5-METHYLINDOLE DERIVATIVES AND ETHYL 7-NITRO-4-METHYLINDOLE-2-CARBOXYLATE

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ABSTRACT

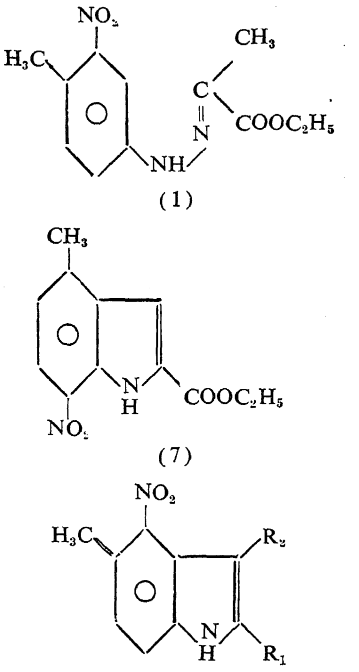
When ethyl pyruvate 4-methyl-3-nitrophenylhydrazone was treated with PPA yielded mainly ethyl 4-nitro-5-methylindole-2-carboxylate (45%) whose structure was confirmed on the basis of PMR data. Similarly, ethyl 7-nitro-4-methylindole-2-carboxylate was obtained in low yield (26%) from the corresponding hydrazone. It has been postulated that the transition state involved in the formation of former might be possessing some stabilising features which lead to higher yield. 4-Nitro-5-methylindole and its 3-aldehyde were also prepared.

THE Fischer indole synthesis[1] is a versatile reaction applied widely for the synthesis of various indole analogues of biological importance from the corresponding arylhydrazones of ketones. But, it is of particular interest to study the indolisation of *m*-substituted phenylhydrazones of ketones[2,3] under the conditions of the Fischer indole sytnthesis, wherein, one expects the formation of 4- and 6-substituted indole analogues. This prompted to report the present work.

Ethyl pyruvate 4-methyl-3-nitrophenylhydrazone (1) was treated with polyphosphoric acid (PPA) to yield mainly ethyl 4-nitro-5-methylindole-2-carboxylate (2) (45%). This was hydrolised with alcoholic KOH to the corresponding acid (3) which was decarboxylated in presence of copper oxide to get 4-nitro-5-methylindole (4). The indole (4) was reacted with phosphorous oxychloride and dimethyl formamide to give indole-3-carboxaldehyde (5) in 85% yield. Similarly, when ethyl pyruvate 5-methyl-2-nitrophenyl-hydrazone (6) was treated with PPA, yielded ethyl 7-nitro-4-methylindole-2-carboxylate (7) (26% yield).

The structure of indolecarboxylate (2) has been established on the basis of PMR data. The location of nitro group at C-4 is evident from the two doublets (J = 8 Hz) shown one at 7·37 δ (C-6 proton) and another at 7·91 δ (C-7 proton). The C-3 proton signal is observed at lower field (7·5 δ) than that (7·22 δ) has been reported[4] earlier for 2-carbethoxyindole in same solvent. The deshielding effect of nitro group is responsible for shifting C-3 proton signal to the lower field by 0·28 ppm and this is an additional proof for the presence of nitro group at C-4. The singlet at 2·74 δ is due to C-7 methyl protons and it is at lower field (deshielded by C-4 NO_2) than the methyl signals (2·4–2·46 δ) reported[5] by us for methylindole analogues having no nitro function. A triplet (1·5 δ, J = 7 Hz) and a quartet (4·53 δ) are due to ester ethyl group.

SCHEME 1



	R_1	R_2
(2)	—$COOC_2H_5$	—H
(3)	—COOH	—H
(4)	—H	—H
(5)	—H	—CHO

During present study, it is noted that the hydrazone (1) gave higher yield of indolic product (2) than the hydrazone (6) and it is in agreement with an earlier report[6]. Similarly, previous studies[2,3] have also indicated the preponderant formation of 4-nitroindole derivative during the indolization of 3-nitrophenylhydrazones of ketones using different acidic cyclising agents. It has been well established[7] that the nitro group is *meta-ortho*-directing in electrophilic aromatic substitution and the yields of *ortho-* and *meta*-products depend to certain extent on the acidity of medium. In view of all these above results, it is felt that apart from the acidity of medium, the transition state involved[8] in the reaction of the Fischer indole synthesis which leads to the 4-nitroindole derivative might be possessing some stabilizing features which might be absent or less important in the transition states leading to the 6-nitroindole derivative or 7-nitroindole derivative.

EXPERIMENTAL

Eihyl pyruvate 4-*methyl*-3-*nitrophenylhydrazone* (1)

This was prepared from 4-methyl-3-nitroaniline (10 g) and ethyl α-methylacetoacetate (10·3 g) according to the procedure described earlier[9] and the hydrazone (1) was recrystallised from alcohol as orange plates (8·5 g), m.p. 122° (Found: C, 54·18; H, 5·82; N, 15·69. $C_{12}H_{15}N_3O_4$ requires C, 54·37; H, 5·66; N, 15·85%).

Ethyl pyruvate 5-*methyl*-2-*nitrophenylhydrazone* (6)

Similarly, this hydrazone (6) was prepared from 5-methyl-2-nitroaniline (10 g) and recrystallised from alcohol to get orange needles (8 g), m.p. 143° (Found: C, 54·62; H, 5·87; N, 15·99. $C_{12}H_{15}N_3O_4$ requires C, 54·37; H, 5·66; N, 15·85%).

Ethyl 4-*nitro*-5-*methylindole*-2-*carboxylate* (2)

A finely powdered hydrazone (1) (10 g) was added at 80° to polyphosphoric acid prepared from phosphorous pentoxide (50 g) and orthophosphoric acid (30 ml) and the mixture was stirred further at 100–105° for 15 min. The mixture was worked out as per literature procedure[3] and the product was repeatedly crystallised from benzene to get yellow microscopic needles (4·3 g), m.p. 209°. TLC [Silica gel G, Benzene-Ethyl acetate (4:1)] Rf. 0·7 [Found: C, 58·24; H, 5·00; N, 11·16. $C_{12}H_{12}N_2O_4$ requires C, 58·06; H, 4·87; N, 11·28%; PMR, CD_3COCD_3 (TMS) ppm 1·5 (3H, *t*, J = 7 Hz, ester CH_3); 4·53 (2H, q, ester-CH_2-); 2·74 (3H, s, C-7 CH_3); 7·37 (1H, d, J = 8 Hz, C-6H); 7·91 (1H, *d*, J = 8 Hz, C-7H); 7 5 (1H, d, J = 2 Hz, C-3H].

Ethyl 7-*nitro*-4-*methylindole*-2-*carboxylate* (7)

The hydrazone (6) (10 g) was added at 100 °C to PPA prepared from P_2O_5 (50 g) and orthophosphoric acid (30 ml) and the mixture was stirred further at 115–120° for 20 min. It was worked as described for (2) and the solid was repeatedly extracted with petroleum ether (60–80°) to yield yellow solid which was recrystallised from petroleum ether to get yellow tiny needles (2 5 g) m.p. 119° (Found: C, 58·17; H, 4·69; N, 11·47. $C_{12}H_{12}N_2O_4$ requires C, 58·06; H, 4·87; N, 11·28%).

4-*Nitro*-5-*methylindole*-2-*carboxylic acid* (3)

A mixture of indole-2-carboxylate (2) (6 g) in ethanol (100 ml) and a solution of KOH (50%, 10 ml) was heated under reflux for 2 hr. and then left at room temperature for 4 hr. It was worked out as described earlier[3] and the product was recrystallised from alcohol to get yellow granules (4·5 g), m.p. 262° (d) (Found: C, 54·61; H, 4·10; N, 13·01. $C_6H_3N_2O_4$ requires C, 54·54; H, 3·66; N, 12·73%).

4-*Nitro*-5-*methylindole* (4)

The indole acid (3) (4 g) was decarboxylated as per literature procedure[3] and the solid on recrystallisation from ethanol gave the desired *indole* (4) as light orange cubes (1·5 g), m.p. 120°. (Found: C, 61·20; H, 4·60; N, 15·45. $C_9H_8N_2O_2$ requires C, 61·35; H, 4·58; N, 15·59%).

4-*Nitro*-5-*methylindole*-3-*carboxaldehyde* (5)

The indole (4) (1 g) was formylated according to literature procedure[10] and the product was recrystallised from alcohol to yield yellow plates (0·95 g), m.p. 214° (Found: C, 59·03; H, 4·03; N, 14·01. $C_{10}H_8N_2O_3$ requires C, 58·83; H, 3·92; N, 13·72%).

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1. *a.* Fischer, E. and Jourdan, F., *Ber.*, 1883, **16**, 2241.
   b. Robinson, B., *Chem. Rev.*, 1963, **63**, 373.
   c. —, *Ibid*, 1969, **69**, 227.
2. Ockendenden, D. W. and Shofield, K., *J. Chem. Soc.*, 1957, pp. 3175.
3. Parmerter, S. M., Cook, A. G. and Dixon, W. B., *J. Am. Chem. Soc.*, 1958, **80**, 4621.
4. Jardine, R. V. and Brown, R. K., *Can. J. Chem. Soc.*, 1963, **41**, 2067.
5. Gadaginamath, G. S. and Siddappa, S., *Indian J. Chem.*, 1975, **13** (12), 1247.
6. Frasca, A. R., *An. Assoc. Quim. Argentina*, 1962, **50**, 1.
7. Hammond, G. S., Modic, F. J. and Hedges, R. M., *J. Am. Chem. Soc.*, 1953, **75**, 1388.
8. Carlin, R. B., *Ibid*, 1952, **74**, 1077.
9. Rydon, H. N. and Siddappa, S., *J. Chem. Soc.* 1951, p. 2462.
10. Smith, G. F., *Ibid.*, 1954, p. 3842.

TONSILS AND OTHER LEUCOCYTOPOIETIC CENTRES IN THE INDIAN FROG *RANA TIGRINA* (DAUD)

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IN spite of the fact that frog has been the 'basic' animal in the teaching of biology and hundreds of frogs are dissected every year in the laboratories, not much work has been done on its anatomy and physiology. An attempt was made by this author last year[6] to describe some special features in the heart of Indian frog *Rana tigrina.* The present paper is aimed to describe the lymphoid and leucocytopoietic centres in the same species of frog. Such centres in this frog are several and have been found to play significant roles in the life of frog in different parts of the year.

Freshly narcotized and formaline preserved frogs of the desired species were procured from and around Lucknow. They were dissected throughout the year taking into consideration both active and hibernation periods. Gum was injected[4] into the lymphatic system through the dorsal cutaneous lymph sinus of frogs narcotized with 5 ml of 1% Urethan solution. The injection increased the colloidal osmotic pressure in the tissue spaces; this prevented the back-suck of the fluid into the venules and hence led to the swelling of the lymph sinuses. This technique helped in observing the lymph sinuses and studying the lymphoid centres. The suspected lymphoid organs were then exposed and their relation to the blood vessels traced out. Grafts from the suspected centres were macerated on the microslides and examined for their nature and contents. Leishman's stain was used to identify the leucocytes.

The lymphoid centres of frog appeared as irregular masses of yellow or orangish colour in the preserved as well as in the freshly narcotized frogs collected from September to following January. In the active period from July to September, the colour was however lighter. It was not uncommon, but unaccountable, in some frogs to find some centres having a grayish tinge after preservation.

Various lymphoid centres may be described thus:

1. *Pharyngeal tonsils.*—A pair of these lymphoid centres were exposed by cutting open the roof of the mouth cavity where they appeared as small diffused lymphoid masses of yellow colour just beneath the bulgings of the eye balls towards the mouth cavity (Fig. 3). These have been suggested to be homologous with the pharyngeal tonsils of mammals[10]. During the active periods of the life of frog these 'tonsils' contained lymphocytes and leucocytes with scattered adipose cells. The blood cells obviously help killing the bacterial and protozoan invaders which might find way into the mouth cavity and lungs with food or air.

2. *Thymus.*—This is no more believed to be an endocrine centre. Variations were found in its size in different frogs. The thymus, one on each side, was found to be located behind the tympanic membrane and under the depressor mandibulae muscle.

3. *Superior and inferior pectoral lymphoid centres.*—These paired and bilaterally symmetrical centres were spotted by their yellow colour in the vicinity of the pectoral girdle, lying between the skin and the muscles. The inferior lymphoid centre was exposed by cutting away the skin from the chest of frog and lifting the pectoral muscles of the girdle to bring in view coracobrachial muscles. This centre consisted of roughly a triangular mass of lymphoid tissue surrounding the junction of the brachial vein with the musculocutaneous vein. For some distance into the fore limb this mass extended around the brachial vein.

The superior lymphoid centre, initially appearing as a large patch of lymphoid tissue in front of the clavicle of each side, was found attached to the skin overlying it. The centre was exposed in full by lifting the pectoral and coracoradial muscles overlying it. This lymphoid mass was associated with the facio-mandibular vein[2] which passes through this centre. The said pectoral lymphoid centres are bathed by the lymph of the large pectoral lymph sinus (Fig. 1).

4. *Deep pectoral lymphoid centres.*—These are located deep in the thoracic cavity close to the sinus venosus on either side and surround the musculocutaneous veins shortly before they join the precavals. The deep centre is reached by removing the coracoid of the pectoral girdle and

tracing it upto the musculocutaneous vein. It is bathed by the large coelomic lymph sinus.

5. *Precardial or median pectoral lymphoid centre.*—This is a prominent bilobed lymphoid massin in front of the heart lying below the omosternum (Fig. 1) and shortly caudal to it. It appeared to be associated directly with the sinus venosus.

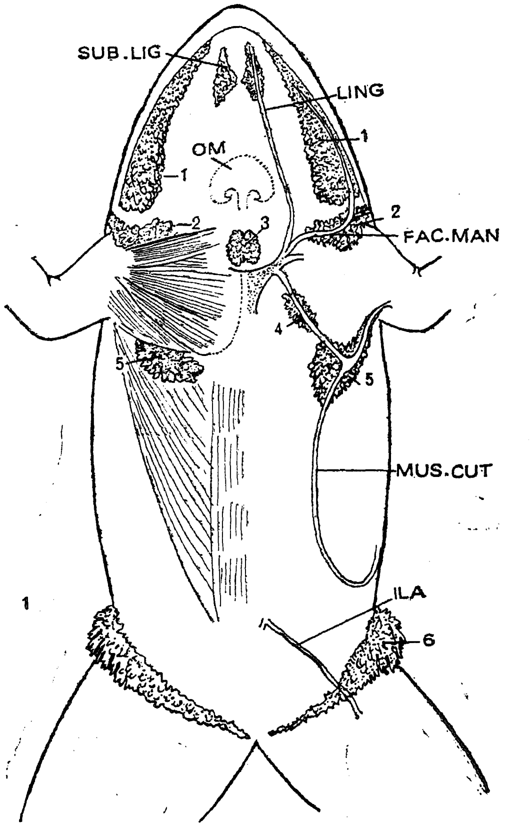

FIG. 1. Diagram of the frog *Rana tigrina* showing the lymphoid centres (ventral) after dissection.

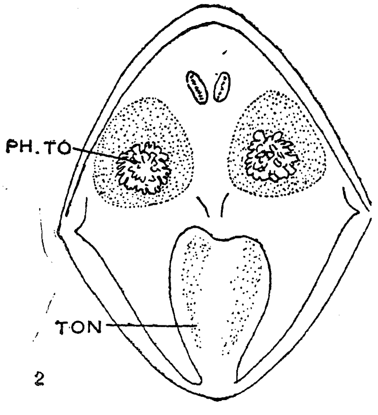

FIG. 2. Diagram of the mouth cavity of *Rana tigrina* showing the pharyngeal tonsils.

6. *Internal mandibular lymphoid centre.*—This paired centre is approached by first cutting away the skin of the throat thereby exposing the large mylohyoid muscles below the floor of the mouth cavity, and further by pulling out the mylohyoid muscles. The internal mandibular centre lies below the mandibular ramus as a long yellowish mass all along the course of the internal mandibular vein. The centre is bathed by the submaxillary lymph sinus.

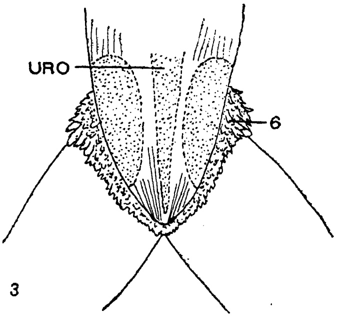

FIG. 3. Diagram of *Rana tigrina* (dorsal) showing the iliac lymphoid centres.

All diagrams in natural sizes.

SUB. LIG. Sublingual tonsils ; LING, Lingual vein ; OM, Omosternum, FAC. MAN, Faciomandibular vein ; MUS. CUT, Musculocutaneous vein, ILA, Iliac vein ; PH. To, Pharyngeal tonsils, TON, Tongue, URO, Urostyle ; 1, Internal mandibular lymphoid centre ; 2, Superior pectoral centre ; 3, Median pectoral centre ; 4, Deep pectoral centre ; 5, Inferior centre ; and 6, Iliac lymphoid centre.

7. *Sublingual lymphoid centre.*—This was exposed without much effort while exposing the internal mandibular centre. Like other centres this too was paired. It appeared as a pair of yellowish elongated masses near the median line and tucked just below the attachment of the tongue. These have been described as sublingual tonsils[5,9].

8. *Iliac lymphoid centre.*—This is a paired centre lying in the abdominal lymph sinus and extending into the dorsal lymph sinus, and by far the biggest. The centres are immediately exposed by cutting away the skin from the pelvic region and the thighs and also the back. Each centre is a frill-like half-girdle around the waist. It begins close to the rectus abdominis muscle on the ventral side and continues towards the back on the side of the coccygeo-iliacus muscle. The iliac centres are associated with the iliac veins.

9. *Dorsal cutaneous lymphoid centres.*—These appear as long and thin yellowish lines on the undersurface of the dorsal body wall and are associated with dorsal cutaneous veins and have direct access to dorsal cutaneous lymph sinus.

DISCUSSION AND CONCLUSION

It is a known fact[7] that there are various masses of lymphoidal tissue in the bodies of amphibia, the more conspicuous are located anterior to the clavicles of frogs or between skin and muscles near the girdles of burrowing species. No such information is in the records for the Indian species of frogs.

From the location of various lymphoid centres described in this paper, it is clear that majority of them are associated with some vein and are placed inside the subcutaneous lymph sinuses. Earlier it was confirmed[1] that during the spring these lymphoid masses produce both lymphocytes and leucocytes but appear to store fat at other seasons. From the fact that lymphocytes are accumulated in the lymphoid centres during active periods of the lives of frogs, some workers[5,9] went to the extent of naming at least some centres, chiefly in the lingual regions as 'tonsils'.

The present author after studying the macerated preparations of all the lymphoid centres, described above throughout the year, found that the findings of workers[5,9] are true for this species of frog as well. During July and August these lymphoid centres showed a marked leucocytopoiesis and were full of leucocytes ; the adipose cells were few. At other times of the year when this species of frog hibernates, the adipose cells gained prominence over the leucocytes and the latter were almost lacking.

Association of lymphoid centres with the veins indicates that the leucocytes produced in the lymphoid centres must be passing on into the blood stream by diapedesis, thus strengthening the body's defence system during the active periods of life of the frog. Since a majority of lymphoid centres are subcutaneous and are bathed by the lymph in the underlying sinuses, their role in the defence of the body from attacks through the skin is quite understandable. Skin being the main source of water during the active periods of frog's life, is liable to bacterial invasion.

It is therefore concluded that the lymphoid centres including the thymus and the tonsils are defence stations from which hosts of phagocytising cells may be mobilized for attack against bacterial and protozoan invaders with food and air reaching the mouth and the lungs, and also those coming with water diffusing into the subcutaneous lymph sinuses during the months of July to September when the frog is most exposed to the aquatic and terrestrial environments. In fact the lymphatic system of this frog could not exist without these lymphoid centres.

The author is gratefull to the management of Lucknow Christian College and specially to its President, for the facilities and encouragement.

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1. Braunmuhl, A., von. "Uber einige myelo-lymphoide und lympho-epitheliale Organe der Anuren," *Zeitschr. Mikr. Anat. Forsch,* 1926, 4, 635.
2. Bhaduri, J. L., Biswas, B. and Das, S. K., "Observations on the course of facial vein and the formation of extrajugular in some Indian frogs and toads," *Jour. As. Soc. Bengal,* 1933.
3. Keynes, G., 'Thymus,' *Ann. Roy. Coll. Surg. Engl.,* 1953, 12, 88.
4. Knower, H. McE., *A Resurvey of the Development of lymphatics and Associated Blood Vessels in Anuron Amphibian Method of Injection,* Wistar Inst. Philadel., 1939, p. 125.
5. Kingsbury, B. F., "Amphibian tonsils," *Anat. Anz.* 1912, 42, 593.
6. Nigam, H. C., *Zoology of Chordates,* 6th Edn. S. Nagin and Co., Jullundur, India, 1973.

—, *Curr. Sci.,* 1975, 44, 810.

7. Noble, G. K., *The Biology of the Amphibia,* Dover Publ., 1954.
8. Moore, J. A., *Physiology of the Amphibia,* Academic Press, 1964.
9. Myers, M. A., *Jour Morph. Physiol.,* 1928, 45, 399.
10. Weichert, C. K., *Anatomy of Chordates,* McGraw-Hill, Co., 1958, p. 233.

LETTERS TO THE EDITOR

MICROWAVE ABSORPTION IN TERNARY MIXTURES OF SOME SUBSTITUTED PIPERIDINES AND XYLENES

SCHALLAMACH[1], Bauer[2] and Magat[3] have shown that the relaxation process for a microscopically homogeneous dipolar mixture involves a single relaxation time. Our earlier[4, 5] investigations for the binary mixtures of piperidines, phenol and substituted benzaldehyde molecules have verified the above view. In the present communication, we have extended this study to some dipolar mixtures of substituted piperidines and Xylenes. The dielectric data and the activation energies of equimolar mixtures of 2-methyl piperidine (I), 3-methyl-piperidine (II) and 3-nitro-*o*-xylene (III) in dilute solutions of benzene have been obtained at four different temperatures. Since the dielectric data for (III) was not available, its relaxation time and thermodynamic parameters have also been determined to facilitate the interpretation of the results.

The relaxation time (τ) has been determined from the fixed frequency method of Gopala Krishna[6] using the experimental technique of Roberts and Von Hippel[7]. The thermodynamical parameters have been calculated using Eyring's[8] theory of Rate process. A.R. chemicals were employed with the usual methods of purification.

It has been observed from Table I that the relaxtion time for a binary mixture decreases with an increase in temperature. This results from the decrease in the viscosity of the solvent with an increase in temperature. The τ values for mixtures are found to be slightly higher than the average of those of its component molecules ($\tau = 6 \cdot 32 \times 10^{-12}$ sec. for 2-methyl-piperidine and $\tau = 7 \cdot 54 \times 10^{-12}$ sec. for 3-methyl-piperidine)[9]. It thus appears that the binary systems show relaxation by co-operation of both the component molecules, influenced more by the molecule which has a higher relaxation time. This is in agreement with the earlier investigations[4, 5].

The value of the relaxation time of 4-nitro-*o*-xylene studied earlier by Mehrotra and Saxena[10] at 22°C is $\tau = 19 \cdot 92 \times 10^{-12}$ sec. Slightly higher value of τ for 3-nitro-*o*-xylene molecule obtained presently can be attributed to the higher intrinsic viscosity or the greater volume swept out for dipole orientation by the molecule.

A quantitative study of the thermodynamical parameters indicates that the system (II + III) has the highest value for molar free energy of activation ($\triangle F_\epsilon$). This is quite expected as the system includes molecules of larger size and thus experiences high intrinsic viscosity in dipole rotation. The free energy of activation ($\triangle F_\epsilon$) is found to be greater than the enthalpy of

TABLE I

Relaxation times and activation energy parameters

Compound	T °K	$\tau \times 10^{12}$ sec	$\triangle F_\epsilon$ KCal/ Mol	$\triangle F_\eta$ KCal/ Mol	$\triangle H_\epsilon$ KCal/ Mol	$\triangle H_\eta$ KCal/ Mol	$\triangle S_\epsilon$ Cal/ Mol	$\triangle S_\eta$ Cal/ Mol
3-nitro-*o*-xylene	290	23·4	2·86	4·22	0·70	2·52	−7·43	−5·85
	298	21·9	2 91	4·27	0·70	2·52	−7·41	−5·88
	308	20·6	2·99	4·34	0·70	2·52	−7·43	−5·92
	318	19·1	3·06	4·40	0·70	2·52	−7.42	−5·93
2-methyl-piperidine + 3-nitro-*o*-xylene	288	20·0	2·74	4·20	0·58	2·52	−7 51	−5·84
	298	18·8	2·82	4·27	0·58	2·52	−7·52	−5·88
	308	17·7	2·90	4·34	0·58	2·52	−7·53	−5·92
3-methyl-piperidine + 3-nitro-*o*-xylene	288	21·4	2·77	4 20	1·19	2 52	−5 48	−5·84
	298	19·3	2·84	4·27	1·19	2·52	−5·52	−5·88
	308	17 1	2·88	4·34	1·19	2·52	−5·48	−5·92
	318	15·3	2·92	4·40	1·19	2·52	−5·45	−5·93
2-methyl-piperidine + 3-methyl-piperidine	288	7 3	2·16	4·20	0·53	2·52	−5·69	−5·84
	298	6·9	2·23	4·27	0·53	2·52	−5·70	−5·88
	308	6·4	2·28	4·34	0·53	2·52	−5·68	−5·92
	318	5·9	2·31	4·40	0·53	2·52	−5·62	−5·93

activation ($\triangle H_e$) resulting in the negative entropy values. This indicates not only a more ordered nature of the activated state but also the co-operation of the nearest neighbours in the process of relaxation, maintaining a relatively higher ordered arrangement.

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1. Schallamach, A., *Trans. Faraday Soc.*, 1946, **43**, 180.
2. Bauer, E., *Ibid.*, 1944, **20**, 37.
3. Magat, M., *J. Chem. Phys.*, 1948, **45**, 93.
4. Dhar, R. L. and Saxena, M. C., *Z. Physik. Chemie, Neue Folge*, 1972, **81**, 189.
5. Shukla, J. P., Dhar, R. L., Gurnani, G. R. and Saxena, M. C., *Ibid.*, 1973, **85**, 76.
6. Gopala Krishna, K. V., *Trans. Faraday Soc.*, 1957, **53**, 767.
7. Roberts, S. and Von Hippel, A., *J. Appl. Phys.*, 1946, **17**, 610.
8. Glasstone, S., Laidler, K. J. and Eyring, H., *The Theory of Rate Process*, McGraw-Hill Book Co., Inc., New York, 1941, p. 548.
9. Singh, B. K. P. N. and Gupta, R. C., *Comm. to Z. Physik. Chemie, Neke Folge*.
10. Mehrotra, N. K. and Saxena, M. C., *Ind. J. Pure and Appl. Phys.*, 1966, **4**, 438.

INFLUENCE OF TEMPERATURE AND CONCENTRATION ON THE ETCHING RATES IN GLASS AND IN PLASTIC*

THE phenomenon of chemical etching on which the solid-state track detectors are finding many applications, is general one and does not disturb the actual physical damage suffered by the material. The whole information which is to be extracted depends upon the measurement of so-called etched track produced by the differential attack of the etchant. Therefore, the precise understanding of the basic etching mechanism is very essential[1] and can be completely described by two different etching parameters:

1. The track etching rate (V) along the radiation damaged region which depends on the energy loss of the particle.

2. The bulk etching rate (v) which refers to an etching vertically to the surface of the sheet and is independent of any energy loss.

The resultant effect of these parameters is the formation of conical etch pits during etching process[2]. Fleischer *et al.*[2] have dealt, in detail, the development of the track in terms of these two parameters. Kinematics of track diameters thus formed has been surveyed by Somogyi *et al*[3]. It is found that these etching rates dod not remain constant during the process of etching. According to Somogyi[4], this process is described by the equation $\tan \alpha = k\, r(t)$ where $r(t)$ is the radius of the etched pit at time t, α is the half of the cone angle of the pit and k which characterizes V is a constant depending upon the energy of the particle, concentration and temperature of the chemical reagent, etc. The differential equation governing this process is

$$dr/dt = v\,[(1 + k^2 r^2)^{\frac{1}{2}} - kr]$$

v and k are connected by semi-empirical relation

$$d^2 = b\; k^{2a-2}\; (vt)^{2a}$$

where d is the diameter of a pit at time t and a and b are numerical constants whose values depend upon the nature of the detector. For glass, according to Somogyi's calculations $a = 1/2$, $b = 4$; the relation is given by

$$d^2 = 4vt/k \text{ not } d^2 = 2vt/k^4.$$

These two etching parameters characterize the track completely and therefore etching has been dealt in the present study. The present work is an attempt to learn the effect of temperature and concentration on these parameters.

Experimental Procedure, Results and Their Discussion

Lexan and microscopic glass slide pieces of size 1 × 1 cm were irradiated to 0·052 MC Cf-252 source^{+} for 20 sec. The samples were etched under controlled conditions of temperature and concentration and measurements were made on diameters of the etched pits. When the tracks were circular or nearly circular, the maximum diameters were measured. In the case of highly elliptical tracks mean of major and minor axes had been computed.

Variation of diameter with etching time at different temperatures and concentrations were studied in detail. The relations thus obtained are linear and not effected by saturation effects which are sometimes observed at larger diameters. The parameter v is easily determined for a given set of conditions. Exponential relation between v and k vs temperaturec can be expressed as $e^{-Q/KT}$ where K is Boltmann's constant, Q and T are the activation energy and temperature (°k) respectively. With the increase of temperature v inincreases and k decreases. For Lexan v is more sensitive to temperature than k while reverse is the case for glass. The results of measurements show that v parameter has more steep dependence on concentration than k indicating that etchants with low concentration are preferred for the best revelation of tracks. For constant temperature and concentrations, v remains practically constant with the increase in etching time while k decreases exponentially with increasing etching time.

CONCLUSIONS

From the analysis of data of different temperatures and concentrations one can choose the etching conditions such that the:

1. Etched tracks with well defined contour are formed.
2. Etching time is within the range of convenience of measurement.

The linear relationships between diameter and etching time give the following best etching conditions:

Detector	Etchant	Concentration	Temperature	Time
1. Lexan	NaOH	6N	65° C	15 min.
2. Glass	HF	20%	35° C	15 sec.

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† The source was purchased from Oak Ridge National Laboratory, U.S.A. and consisted of 1/2″ diameter electrodeposit of 0·1 μ gm Californium oxalate on 1″ platinum disc.

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1. Nand Lal, Saini, H. S. and Nagpaul, K. K., *Radiation Effects*, 1976 (To be published).
2. Fleischer, R. L., Price, P. B. and Walker, R. M., *Nuclear Tracks in Solids*, University of California Press, Berkeley, 1975.
3. Somogyi, G. and Szalay, S. A., *Nucl. Instr. Meth.*, 1973, **109**, 211.
4. —, *Ibid.*, 1966, **42**, 312.

IRON (III) THIOCYANATE COMPLEXES WITH SOME SCHIFF BASES

THE chloro and the bromo complexes of iron (III) with ethylenediamine-salicylaldehyde are reported to be dimeric[1]. A few cyanide complexes of iron (III) with Schiff bases showing interesting magnetic behaviour have been reported recently[2]. We present here the results of our studies on iron (III) thiocyanate complexes of the Schiff bases derived from salicylaldehyde and aniline (salanil), ethylenediamine and 2-hydroxy acetophenone (aphen) or 2-hydroxy 5-methyl acetophenone (maphen), and glyoxal and *o*-aminophenol (glyoxa anil).

2-OH-acetophenone and 2-OH, 5-CH_3-acetophenone were prepared by known methods[3]. The Schiff bases were prepared by condensing the respective aldehyde or ketone with the amines in ethanol. The ligands separated were filtered, washed with ethanol and recrystallised from benzene. An aqueous solution of iron (III) thiocyanate was prepared by mixing 30 ml of $FeCl_3 \cdot 6H_2O$ solution (10%) with 70 ml KCNS solution (10%). The deep red iron (III) thiocyanate was extracted into an ether layer (75 ml). The ether layer separated was then mixed with aphen or maphen in benzene (1 : 1). The salanil and glyoxal bis (2-hydroxy anil) complexes were prepared by refluxing with iron (III) thiocyanate solution in ethanol and methanol respectively ($\sim$4 hrs.). The pH was adjusted to 5 using HAc–NaAc buffer. The dark brown complexes separated were filtered, washed with water and benzene, then with ether and dried in vacuum over P_2O_5.

The analytical data, conductance values, magnetic moments and important infrared frequencies are summarised in Table I. The electrical conductance data of the complexes indicate that they behave as non-electrolytes in nitrobenzene. The decrease in the magnetic moment values from the spin-only value found in the thiocyanate complexes are quite comparable to that of the chloro complexes of ensal[4]. This is due to considerable interaction between iron atoms.

The complexes have been further characterised on the basis of their infrared spectra (KBr and Nujol mull). The broad, medium intensity band observed around 3000 cm^{-1} in the spectra of the free ligands is assignable to the hydrogen bonded–OH group. In the spectra of the aphen and maphen complexes, a broad band appears at 3450 cm^{-1} showing the presence of free hydroxyl group. Thus, the infrared spectra of the complexes Fe (aphen) $(SCN)_2$, Fe (maphen) $(SCN)_2$, along with the analytical data indicate that one of the hydrogens of the hydroxyl groups of the free ligand is replaced by the metal while the other remains free. This type of behaviour is not uncommon in the case of Schiff base complexes[5]. In the case of the other two complexes, Fe (salanil) SCN and Fe (glyoxalanil) SCN, the $-OH$ group of the free ligand disappears on complexation showing that both the hydrogens are replaced by the metal. The free ligand show medium intensity band around 1500 cm^{-1} which is characteristic of the $C = N$ group. This band is shifted to around 1540 cm^{-1} indicating the participation of $C = N$ group in complexes. A strong, sharp characteristic absorption at 2090 and 2120 cm^{-1} observed in the thiocyanate complexes, but absent in the free ligands can be assigned to the thiocyanate group coordinated through nitrogen[6].

TABLE I

Analytical and physical data

Complex	Colour	Fe % Found	Fe % Calcd.	S % Found	S % Calcd.	μ_{eff} B.M	Electrical conductance ohm^{-1} cm^2 mole-1 (nitrobenzene)	IR frequencies and their assignments Cm^{-1}	Assignments
Aphen	Yellow	..	..	..	..	..	..	2900 bm 1510 m	νOH νC = N
Fe (aphen) (SCN)$_2$	Dark brown	11·87	11·96	13·41	13·70	5·37	3·7	3450 m 1540 m 880 m	νOH νC=N νC - S
Maphen	Yellow	..	..	..	..	..	..	2950 bm 1500 m	νOH νC=N
Fe (maphen) (SCN)$_2$	Dark brown	10·95	11·28	12·28	12·93	5·42	9·2	3450 m 1520 m 830 m	νOH νC=N νC–S
Salanil	Yellow	..	..	..	..	..	..	3000 bm 1460 m	νOH νC=N
Fe (salanil)$_2$ SCN	Dark brown	11·18	11·04	6·96	6·32	5·20	3·7	1480 m 860 m	νC=N νC - S
Gly. anil	Colourless	..	..	..	..	..	..	3450 m 1500 m	νOH νC=N
Fe (glyanil) SCN	Dark brown	16 14	15·87	10·30	9·09	3·93	0·6	1520 m 860 m	νC=N νC–S

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---

1. Gerloch, M. and Mabbs, F. E., *J. Chem. Soc., A*, 1967, p. 1900.
2. Nishida, Y., Oshio, S., and Kida, S., *Chemistry Letters*, 1975 (January), p. 79.
3. *Org. Synthesis, Collective Vol.* 4, 1963, p. 106.
4. Bancroft, G. M. and Platt, R. H., *Advances in Inorganic Chemistry and Radiochemistry.* (Emeleus, H. J. and Sharpe, A. G., eds., Academic Press, New York), 1972, **15**, 214 and references therein.
5. Zimmerman, D. N. and Hall, J. L., *Inorganic Chemistry*, 1973, **12**, 2616.
6. Nakamoto, K., *Infrared Spectra of Inorganic and Coordination Compounds*, John Wiley and Sons, Inc., New York, London, 1963.

ANALYTICAL APPLICATIONS OF 6-AMINO-5-NITROSO-2, 4-PYRIMIDINEDIOL

2-AMINO-5-nitroso-4, 6-pyrimidinediol has been used earlier[1] as reagent for Fe^{2+}, CO^{2+} and Ru^{3+}. To investigate the effect of substituents in the molecule, the complexation reactions of the ligand with groups interchanged at C$_2$ and C$_6$ compared to its precursor are now reported. Moreover, the present compound, *viz.*, 6-amino-5-nitroso-2, 4-pyrimidinediol (monosodium salt) is more water-soluble.

The aqueous solution of the ligand is light red in colour absorbing maximum at 225 and 315 nm. The characteristics of its metal complexes are summarised in Table I.

Procedure.—To a suitable aliquot of metal solution (Fe^{2+}, Co^{2+} or Ru^{3+}), sufficient excess of the diol (0·008 M) is added followed by 5·0 ml of acetate buffer (pH 5·0). In the case of Ru^{3+}, 0·1 M NaClO$_4$ strength is also maintained and the solution heated for 45 minutes on a steam bath. After making up the volume, the absorbance is measured at λ_{max} of the complex. The amount of the metal is calculated by comparing the absorbance with a calibrated curve.

TABLE I

Properties of metal complexes with the diol

Property	Fe (II)-complex	Co (II)-complex	Ru (III)-complex
Colour	Blue	Yellow	Pink
λ_{max} (nm)	610; 650	380	510
ϵ_{max} ($1 \cdot mole^{-1} \cdot cm^{-1}$)	16,000 at 650	14,600	22,500
Diol needed for full colour development	90-Molar concentration	50-Molar concentration	30-Molar concentration
pH range to attain maximum absorbance	4·3–6·3	2·7–7·6	3·5–6·5
Molar composition (metal:diol)	1:3 (at 610 nm) 1:4 (at 650 nm)	1 : 3	1 : 3
Beer's law (ppm)	Upto 1·6	Upto 1·8	Upto 3·0
Sandell's sensitivity	0·0033	0·0040	0·0044

In determination of 1·4 ppm of iron, the following ions could be tolerated to the limits given in parentheses (ppm): Cl^-, Br^-, SCN^-, SO_4^{2-} or phthalate (1,000); Cd^{2+} (500); thiourea or Tl^+ (100); Mn^{2+} (75); I^-, NO_3^-, Ag^+, Zn^{2+} (50); F^-, $S_2O_3^{2-}$, PO_4^{3-}, Ca^{2+}, Sr^{2+} or Ba^{2+} (20); Ni^{2+} (2). Serious interference was observed with NO_2^-, $C_2O_4^{2-}$, EDTA, citrate, BO_3^{3-}, Cu^{2+}, Au^{3+}, Hg^{2+}, Fe^{3+}, Co^{2+} and Ru^{3+}.

In determination of 1·5 ppm of cobalt, the following ions could be tolerated: Cl^-, Br^-, SCN^- or phthalate (1,000); I^-, SO_3^{2-} or thiourea (400); $C_2O_4^{2-}$, $C_4H_4O_6^{2-}$, PO_4^{3-} or citrate (20); NO_3^-, Ag^+, Zn^{2+} or Tl^+ (50); Cd^{2+}, Pb^{2+} or V^{4+} (40); Ca^{2+}, Sr^{2+} or Ba^{2+} (20). NO_2^-, EDTA, BO_3^{3-}, Cu^{2+}, Au^{3+}, Sn^{4+}, Fe^{2+}, Fe^{3+}, Ni^{2+} and Ru^{3+} interfere.

In determination of 2·0 ppm of ruthenium, the following ions were tolerated: Cl^-, Br^-, I^- or CH_3COO^- (1,000); NO_3^- (500); $C_2O_4^{2-}$ (100); SO_4^{2-}, $C_4H_4O_6^{2-}$, PO_4^{3-} or citrate (50); F^-, SCN^- Cd^{2+}, Pb^{2+}, V^{4+} or UO_2^{2+} (10). NO_2^-, $S_2O_3^{2-}$, thiourea, EDTA, BO_3^{3-}, Cu^{2+}, Sn^{2+}, Fe^{3+}, CO^{2+} and Ni^{2+} interfere.

Comparing the data of the present compound with that of the earlier[1], it is evident that this compound affords better sensitivity and selectivity, provides wider pH range for the study and lower concentration is required for the complexation and determination of ruthenium. However, for iron (II) determination, it is comparatively inferior.

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---

1. Kundra, S. K., Katyal, M. and Singh, R. P., *Curr. Sci.*, 1975, **44**, 81.

TERPENOIDS OF *PTEROCARPUS SANTALINU* LEAVES

IN our earlier publications we have reported the terpenoids of *Pterocarpus santalinus* heartwood[1], sapwood[2] and the bark[3-4]. The heartwood was found especially rich in sesquiterpenes and contained six of them besides two triterpenes. On going from inner heartwood to the outer sapwood and bark an increase in triterpene composition was noticed. The sapwood had four triterpenes and only one sesquiterpene while the outermost bark was found to certain nine triterpenoids but no sesquiterpene. In order to study further the qualitative variations in the chemical components of the various parts of the tree, the leaves of *Pterocarpus santalinus* were next investigated.

The air dried leaves obtained from Waltair were powdered and extracted successively by boiling with (*i*) petroleum ether (60–80°, 3 × 5 hr), (*ii*) benzene (3 × 4 hr.), (*iii*) chloroform (3 × 5 hr) and (iv) ethanol (3 × 4 hr). Petroleum ether and benzene extracts were found very similar on T.L.C. and mixed together.

Petroleum ether and benzene extracts : The concentrate of the combined extractives on column chromatography over silica gel gave four fractions (I), (II), (III) and (IV).

Fraction I : It required further chromatography over Ag^+/SiO_2 (6%) in order to separate the compounds A and B which otherwise did not resolve on silica gel alone. Both the compounds crystallized from alcohol as colourless needles, m.p. 178° and 172° respectively and gave + ve LB test. They were identified as β-amyrone and lupenone respectively on the basis of their spectral and physical properties and the identities confirmed by direct comparison with respective authentic samples.

Fraction II : On Crystallization from alcohol it afforded pure compound C as colourless needles, m.p. 206° and gave +ve LB test. The compound was identified as epi-lupeol since on Jones' oxidation it afforded lupenone and was itself different from lupeol.

Fraction III : It was chromatographed over Ag^+/SiO_2 to separate compounds D and E which crystallized from alcohol as needles, m.p. 199° and 212° respectively. They were identified as β-amyrin and lupeol respectively on the basis of their physical properties and spectral data.

Fraction IV : Column chromatography over Ag^+/SiO_2 separated compounds F and G, m.p. 138° and 169° respectively. They gave green colour in LB test and were identified as sitosterol and stigmasterol and the identity confirmed by direct comparison with respective authentic samples.

Chloroform extract : The extract was subjected to column chromatography over silica gel which yielded compounds H and I in addition to the small amounts of aforesaid compounds. The compounds crystallized from alcohol as needles, m.p. 238° and 258° respectively and gave +ve LB test. They were identified as erythrodiol and betulin on the basis of their spectral data and confirmed by direct comparison with respective authentic samples.

Alcohol extract: It contained leucoanthocyanins and the anthocyanidin produced by heating with alcohol hydrochloric acid was found to be cyanidin by direct comparison by paper chromatography. The leuco compound appeared to be highly polymeric and could not be studied further.

The occurrence of the compounds of lupene series and the corresponding compounds of biogenetically related oleanane series as pairs[1] separable by Ag^+/SiO_2 column chromatography only is interesting.

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1. Kumar, N., Ravindranath, B. and Seshadri, T. R., *Phytochemistry*, 1974, 13, 633.
2. — and Seshadri, T. R., *Curr. Sci.*, 1974, 19, 911.
3. — and —, *Phytochemistry*, 1975, 14, 521.
4. — and — (Under publication).

CHEMICAL CONSTITUENTS OF *UMBILICARIA INDICA* FRÉY AND *RAMALINA FARINACEA* L. ACH.

THE lichen *Umbilicaria indica* Frèy (Umbilicariaceae) was collected from the rocks near Govindaghat in the Chamoli District of the Western Himalayas. A survey of literature showed that the lichen was not previously studied for its chemical components. The dark brown coloured lichen (15 g) with dark pustules was dried, powdered and extracted with light petroleum ether (b.p. 60–80°), ether and acetone in succession. Traces of wax was found in the light petroleum ether extract. The ether and acetone extracts contained the same phenolic constituents (TLC) and therefore were mixed and evaporated to dryness. The residue was treated with water and the water insoluble part (0·18 g) was found to be a mixture of two phenolic components by TLC[1] (Silica gel-G ; benzene : dioxan : acetic acid, 90 : 25 : 4). Their R_f values corresponded to those of lecanoric and gyrophoric acids. They were separated by fractionation from acetone. The major component of the mixture was shown (UV, IR and TLC) to be lecarnoric acid, m.p. and mixed m.p. with an authentic sample of lecanoric acid, 174–6°. The minor component was shown (UV, IR and TLC) to be gyrophoric acid, m.p. and mixed m.p. with an authentic sample, 220° d. The water soluble portion of the acetone extract was treated with Norit and the aqueous solution filtered. On concentration it gave a small amount of residue having mannitol (paper chromatography)[2].

Ramalina farinacea L. Ach. (Usneaceae) was collected from trees near Eturu Nagaram in Warangal District, India. Survey of literature[3] showed that various samples of this lichen yielded divergent results. In the present case 50 g of the powdered lichen was extracted as in the previous case. The light petroleum extract yielded (+)-Usnic acid, m.p. 203–4° ; $[\alpha]_D$ + 493° (*c*, 1·0 in chloroform) (0·15 g, 0·3%) identified by a direct comparison with an authentic sample. The ether extract yielded a colourless compound (0·1 g., 0·2%) which crystallised from 80% acetone as needles, m.p. 172–3°. Its homogeneity was revealed by TLC. The acetone extract was evaporated to dryness and the residue (1·5 g) extracted with hot water. The water insoluble residue revealed the presence of two phenolic compounds by TLC, the one with the higher R_f value being identical with the compound obtained from the ether extract. The solid was repeatedly washed with ice-cold acetone. The acetone solution on evaporation gave a residue (1·2 g, 2·4%) which on repeated recrystallisation from 80% acetone gave colourless needles, m.p. 172–3°. The compound was identical with the ether compound (Found : C, 61·2 ; H, 5·2 : Calc. for $C_{17}H_{16}O_7$: C, 61·4 ; H, 4·8%). Its colour reactions and spectral data[4,5] (UV, IR and NMR) indicated the compound to be evernic acid. It was subjected to menthanolysis with 5% methanolic KOH at 40° C in nitrogen atmosphere for 2 hours and the acid and the ester components isolated as usual. The methanolysis products were identified (m.p., mixed m.p., TLC, UV, IR and elemental analysis) as methyl everninate and orsellenic acid. This confirmed the identity of the compound as evernic acid.

The acetone insoluble residue (0·1 g, 0·2%) was recrystallised from 80% acetone to obtain wooly needles, m.p. 198° (Found : C, 62·1 ; H.5·6 : Calc. for $C_{18}H_{18}O_7$: C, 62·4 ; H, 5·2%). Light absorption in ethanol : max. at 275 and 306 nm (log ϵ : 4·33 and 4·03. Main IR bands : ν_{max}^{nujol} 3509 (phenolic OH), 1667 (chelated depside carbonyl), 1639 (chelated carboxy carbonyl) cm^{-1}. It gave a violet ferric reaction and a positive homofluorescein reaction. It formed an yellow solution with conc. HNO_3 and a pale pink solution on standing with alkali. These properties indicated that the compound might be obtusatic acid. The methyl ester of the acid (brief treatment with diazomethane in ether solution) crystallised from methanol as colourless needles, m.p. 175° (Lit. m.p. for methyl obtusate[6], 175°) and acetylation of the acid (acetic anhydride–H_2SO_4) gave the acetyl derivative which separated from ethyl acetate–light petroleum ether as prisms, m.p. 172–4[6] (Lit.[6] m.p. 175°). Its methyl ester dimethyl ether (diazomethane–methanol/ether ; 48 hr) crystallised from methanol as colourless needles, m.p. 126–127° (Lit.[6] m.p. 127°) (Found : C, 64·7 ; H, 6·4 : Calc. for $C_{21}H_{24}O_7$: C, 64·9 ; H, 6·2%). The NMR spectral data of the methyl ester dimethyl ether clearly indicated the various protons of the different functional groups. Comparison with an authentic sample of obtusatic acid[7] confirmed their identity. Incidentally this is the first report of the occurrence of obtusatic acid in the Indian lichens.

The water soluble portion of the acetone extract was treated with Norit, filtered and the filtrate evaporated. The residue (0·05 g., 0·1%) was triturated with cold absolute methanol and filtered. The residue crystallised from methanol–absolute ether as colourless crystals, m.p. 102°. It was identified as D-arabitol by paper chromatography[2] and by a direct comparison with an authentic sample.

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---

1. Ramaut, J. L., *Bull. Soc. Chim. Belg.*, 1963, 72, 316.
2. Godin, P., *Nature, Lond.*, 1954, 174, 134.
3. Culberson, C. F., *Chemical and Botanical Guide to Lichen Products*, The University of North Carolina Press, Chapel Hill, 1969, p. 493.
4. Rao, P. S., Sarma, K. G. and Seshadri, T. R., *Proc. Ind. Acad. Sci.*, 1967, **66 A**, 1.
5. Huneck, S. and Linscheid, P., *Z. Naturforschg.*, 1968, 23 B, 717.
6. Asahina, Y. and Shibata, S., *Chemistry of Lichen Substances*, Japan Society for the Promotion of Science, Ueno, Tokyo, Japan, 1954, p. 70.
7. Kindly supplied to us by Prof. S. Huneck.

CHEMICAL EXAMINATION OF LEAVES OF *BARRINGTONIA ACUTANGULA* GAERTN.

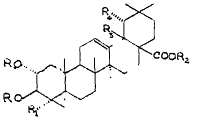
Several polyhydroxy triterpenes were isolated in the past from the various parts of *Barringtonia acutangula* Gaertn[1-4]. We now describe the isolation and structural elucidation of a new trihydroxy triterpene monocarboxylic acid, now named acutangulic acid, from its leaves, in which it is accompanied by stigmasterol-β-D-glucoside and barringtogenic acid (I).

Acutangulic acid (II) $C_{30}H_{48}O_5$ m.p. 290–1°, $[\alpha]_D$ + 15°, diacetate (III) m.p. 188–9° $[\alpha]_D$ − 35°, methyl ester diacetate (IV) m.p. 219–20°, has a *trans* glycolic function (1 mole of periodate in 4–6 hrs.) which can be located at 2 α, 3 β from its NMR spectrum. The hydrogen on the 2 α-acetoxyl appeared as an ill-defined quartet at δ 5·05, 5·12, 5·20, 5·28 J 4, 9 Hz and the proton on 3 β-acetoxyl as a doublet at δ 4·8, 4·9 J 9 Hz. This is in close agreement with a number of 2 α, 3 β-dihydroxy triterpenes occurring in nature[5].

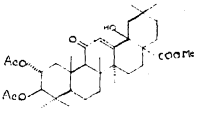
The third hydroxyl could not be acetylated even under forced conditions (with perchloric acid) and diacetyl methyl acutangulate exhibited a singlet resonance at δ 2·6 in its NMR spectrum indicating its tertiary character. It is further supported by its unusual resistance towards pyridine-CrO_3 or CrO_3–AcOH. But with the latter reagent after prolonged reaction, an α : β-unsaturated ketone (V) was obtained, m.p. 165° IR 3540 (–OH) and 1650 cm^{-1}. Obviously, the ketone could be 11-keto $\Delta^{12\,:\,13}$ methyl diacetylacutangulate.

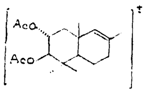
Mass spectral fragmentation[6] of diacetylacutangulic acid as well as its methyl ester furnished the expected RDA fragments (VI, VII and VIII) suggesting that the hindered hydroxyl might be in D or E ring and not in A or B. The location of this hydroxyl is revealed by its ready formation of methylanhydrodiacetylacutangulate (IX) m.p. 180–2°, $[\alpha]_D$ + 116° with $POCl_3$–Py. at reflux temperature or even glacial AcOH-sulphuric acid at reflux temperature for 4 hrs. This anhydro-ester (IX) contained a S *cis*-diene system (U.V. 246, 252 nm log ε 4·01 and 4·02 respectively), the new double bond conjugating with 12 : 13-double bond.

The NMR spectrum of the methylanhydrodiacetylacutangulate (IX) contained an additional proton at δ 5·43 d (J 1 Hz) in addition to the common 12 C–*H*, which appeared as a multiplet at δ 5·50. This negligible coupling constant for the new olefenic proton suggests a long-range coupling in the molecule. These experiments indicate that the tertiary hydroxyl could be located only at C–18. If it were so, the newly introduced double bond must occupy 18–19 positions and could be identical with methyl anhydrodiacetylarjunate, obtained by similar dehydration from methyldiacetyl arjunate (XI), a derivative of arjunic acid (X)[5]. Comparison with an authentic sample established its identity unequivocally (mmp and IR).

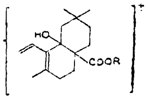


I $R=R_2=R_3=R_4=H$; $R_1=COOH$
II $R=R_2=R_4=H$; $R_1=CH_3$; $R_3=OH$
III $R=Ac$; $R_1=CH_3$; $R_2=R_4=H$; $R_3=OH$
IV $R=Ac$; $R_1=R_2=CH_3$; $R_4=H$; $R_3=OH$
X $R=R_2=R_3=H$; $R_1=CH_3$; $R_4=OH$
XI $R=Ac$; $R_1=R_2=CH_3$; $R_3=H$; $R_4=OH$

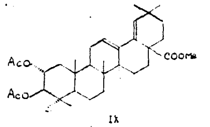




VI m/e 308



VII R=H m/e 264
VIII R=Me " 278



IX

In an attempt to determine the steric configuration of the 18–hydroxyl, information was sought through lactonisation under normal conditions as in oleanolic acid[7]. Acutangulic acid (II) always afforded a mixture of compounds which could not be resolved. If the hydroxyl is 18 α-situated, lactonisation would be easier and a single product may be expected. Since a mixture is obtained it is presumed that D/E rings are *cis*- fused as in oleanolic acid and 18–OH is β-oriented.

On this basis acutangulic acid is tentatively assigned the structure 2 α, 3 β, 18 β-trihydroxy-olean-12 en-28-oic acid (II) and happens to be the first recorded instance of an 18 hydroxy pentacyclic triterpene occurring in nature, although 20 α-hydroxy tetracyclic triterpenes and steroids (steroidal numbering) have been known for quite some time. Their origin must have been from a common non-classical carbonium ion intermediate during squalene cyclisation[8] which is believed to be responsible for the structural variation well known in rings D, E or the side-chain of triterpenes or steroids.

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1. Barua, A. K., Maiti, P. C. and Chakraborti, S. K., *J. Am. Pharm. Assoc.*, 1961, **50**, 937.
2. — and Chakraborti, P., *Tetrahedron*, 1965, **21**, 38.
3. Prakasa Sastry, C. S. and Ramachandra Row, L., *Ibid.*, 1967, **23**, 3787.
4. Barua, A. K., Patel, S. K. and Dutta, S. P., *Sci. and Cult.*, 1968, **34**, 259.
5. Ramachandra Row, L., Suryanarayana Murty, P., Subba Rao, G. S. R., Prakasa Sastry, C. S. and Rao, K. V. J., *Ind. J. Chem.*, 1970, **8**, 716.
6. Budzikiewicz, H., Wilson, J. M. and Carl Djerassi, *J. Am. Chem. Soc.*, 1963, **85**, 3688.
7. Barton, D. H. R. and Holness, N. J., *J. Chem. Soc.*, 1952, p. 78.
8. Connolly, J. D. and Overton, K. H., In : *Chemistry of Terpenes and Terpenoids*, Edited by A. A. Newman Academic Press, 1972, Chapter 5, p. 221.

OESTROGEN INJECTION TEST FOR EARLY PREGNANCY DIAGNOSIS IN MARES*

Review of Litreature

TSUNEKANE *et al.* (1953) reported failure of heat symptoms in pregnant cows with subcutaneous injection of 100,000 i.u. of oestrogen one to three days before next expected oestrum. Nishikawa (1959) observed that with subcutaneous administration of 2–3 mg of stilbestrol diproprionate on 16th to 17th day following last service, pregnant mares failed to return to oestrum; and non-pregnant showed oestrous symptoms within two to three days. The efficacy of the test was zero between two to twelve days, 50% on 13th day, 67% on 14th day, 86% on 15th day and 100% from 16th day onwards from last service. With combined administration of oestrogen and testostrone in sows the efficacy of the test claimed was 100% by Jochle and Schilling (1965) and 98% by Rothe (1971).

Matcrials and Methods

The present study was conducted at a thorough bred equine breeding station with 98 mares and six stallions. Thirty mares were selected at random for the study. Duration of oestrum of individual mares was recorded for a period of three months prior to the commencement of breeding season, by a study of their behavioural symptoms to teaser stallions or mares at paddocks, and by rectovaginal examination.

Two or three mg of stilbestrol diproprionate was injected subcutaneously on 16th to 17th day following last service and onset of oestrum investigated. Results recorded as pregnant were compared with those of rectal palpation and biological tests between 45 to 60 days.

False positive reactors were induced in oestrum by intrauterine infusion of about 1000 ml of normal saline with antibiotics at 37·5° C.

Results

Out of thirty mares tested 24% failed to exhibit oestrum following oestrogen injection during the first oestrum, 35% during the second, 18% during the third, 18% during the fourth and 6% during the fifth. The interval between the test injection and onset of oestrum was one day in 20%, two days in 20%, three days in 50% and six days in 10% mares. The efficacy of the test was 90%. Only three mares showed false positive reactions consistently throughout the breeding season.

Discussion

The oestrogen injection test in mares is based on the principle that the effect of administration of subclinical doses of oestrogen is related to the functional status of corpus luteum. Nishikawa (1959) reported that the incidence of induced oestrum in non-pregnant mares with administration of oestrogen varied from 0% between two to twelve days to 100% on 16th to 17th day after last service. In pregnant mares, on the other hand, the functional status of gravid corpus luteum remained undisturbed with the absence of oestrous symptoms. In the present study the efficacy of the test was 90% when compared with biological tests.

Repeated false reactions in three mares during the entire breeding season are probably due to early embryonic death with the resorption of foetus and persistence of corpus luteum. The incidence of early embryonic deaths in mares has been reported to be 10 to 30% by Britton (1947), and resorption of foetus resulting from corpus luteum insufficiency has been recorded between 42 to 100 days by Sagar (1962). Nishikawa (1959) observed false reactions in mares with prolonged oestrum and attributed them to persistency of corpus luteum.

False reaction in human patients to early pregnancy diagnosis with oestrogen-progesterone therapy has been attributed to persistency or cystic conditions of corpus luteum by Graber *et al.* (1970).

The fertility of the mares following the test dose of oestrogen remained unaffected. Development of normal follicles and incidence of 35% pregnancies following coitus during second, 18% each during third and fourth and 6% during fifth induced oestrums in the tested mares probably indicates absence of any adverse effects on ovarian activity. In equine breeding programmes this test seems to provide an efficient technique for confirmation of early pregnancy and for forced induction of a probable normal oestrum in non-pregnant mares thus providing repeated services in limited breeding season for obtaining highest conception rates.

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1. Britton, J. W., *Corn. Vet.*, 1947, **37**, 14.
2. Graber, E. A., Barber, H. R. K. and C. Rourke, J. J., *Am. J. Obstet. Gynaec.*, 1970, **108**, 991.
3. Jochle, W. and Schilling, E., *J. Reprod. Fert.*, 1965, **10**, 439.
4. Nishikawa, Y., *Studies on Reproduction in Horses*, Edn. 1. Japan Racing Association, Tokyo, Japan 1959.
5. Roberts, S. J., *Veterinary Obstetrics and Genital, Diseases*, 2nd Edn., 1972, p. 529, Published by author.
6. Rothe, K., *Dt. Tierazti. Wachr.*, 1971, **78** (*Vet. Bull.* **41**, 6526).
7. Sagar, F. C., (1962), Cited by Roberts (1972),
8. Tsunekane, T., Terade, K. and Watanabe, M., *Exp. Rep. Govt. Exp. Sta. Animal Hyg. Tokyo.* 1953, No. **26**, 135 (*Vet. Bull.*, **24**, 2942).

TRANSFER OF HILY CHARACTER TO INDIAN BARLEY

MOST of the commercially grown Indian barleys are deficient in laysine and threonine and are, therefore, of nutritionally poor quality[1]. Hence, use was made of the high lysine, high protein genotype, Hiproly[2], to breed barley lines with improved nutritional quality. Some lines obtained from the cross between Jyoti and Hiproly, and Vijaya and Hiproly have been identified recently[3]. This communication presents additional information which confirms the successful transfer of the Hily character of Hiproly for the first time to an agronomically superior Indian barley, Jyoti.

The barley line, 1098–2 (in the F_7 generation) and the parents, Jyoti and Hiproly, were grown in the experimental fields of I.A.R.I. during the *rabi* season of 1974–75 under normal fertility levels of N, P and K (30 : 20 : 20 kg/ha). Ten individual plants of line 1098–2 and Jycti were harvested separately to obtain data on the number of spikes, total weight of the grains, weight of the grains per spike, thousand kernel weight and the number of grains per spike.

Protein was determined in the samples by the micro-Kjeldahl method[4] and the amino acid analysis was carried out on the acid hydrolysate of the sample using a Technicon Sequential Multisample Amino Acid Analyser[5]. Tryptophan was determined colorimetrically[6].

Chemical score was calculated as described earlier[6] by the method of Block and Mitchell[7], using the reference pattern suggested recently by the FAO[8].

A comparison of the content of essential amino acids in the newly developed line, 1098–2, with that in the parent, Jyoti, showed that there was an enhancement of these amino acids in 1098–2 (Table I). The increase in the content of lysine in particular was reflected by a marked improvement in the chemical score of the grain protein of the line 1098–2 over that of Jyoti (Table I).

The grain protein content of the line 1098–2 was higher than that of Jyoti (Table I). On 100 g whole meal basis the line provides 0·646 g lysine as compared to 0·414 and 0·838 by Jyoti and Hiproly, respectively.

The Hily gene of Hiproly, which seems to be transmitted as a simple Mendelian character, brings about a change in the endosperm protein thus increasing the content of lysine, threonine, methionine and aspartic acid[9]. The content of these amino acids was considerably enhanced (Table II). It is of interest to note that these amino acids belong to the aspartate family. While it is obvious that the Hily gene brings about a change in the content of these amino acids by acting at the biosynthetic level, studies need to be carried out to elucidate the metabolic controls involved. That the shrivelled grain character of Hiproly is not pleiotropic with the Hily gene was suggested earlier[10]. From the present study it becomes obvious that Hily gene can be combined with well-filled grains (Table III). From the data

TABLE I

Content of essential amino acids, grain protein and chemical score of the developed line as compared to that of its parent

Amino acid	Variety/Strain		
	1098–2	Jyoti	Hiproly
Lysine	4·04	3·05	4·07
Histidine	2·49	2·30	2·80
Arginine	5·59	5·29	6·19
Threonine	3 02	2 67	3·00
Valine	4·89	4·60	4·64
Methionine	2·10	1·80	2·46
Isoleucine	3·79	3·62	3·27
Leucine	6·72	6·53	6·81
Pehnylalanine	5·39	4·98	5·01
Tryptophan	1·18	1·10	1·21
Protein % N × 6·25	16·0	13·6	20 6
Chemical score	73	55	74

TABLE II

Increase in the amino acids characterised by the Hily gene in the grain protein of 1098–2 *over that of* Jyoti.

	Lysine	Threonine	Methionine	Aspartic acid
Jyoti	100	100	100	100
10 98–2	132	113	117	155

TABLE III

Thousand kernel weight (g) of the developed line compared to that of the parents over three seasons

Strain/Variety	1973	1974	1975	Mean
1098–2	36	36	35	35·7
Jyoti	38	39	39	38·7
Hiproly	27	28	28	27·6

TABLE IV

Cumulative data on the ancillary characters of 1908–2 *and Jyoti*

Strain/Variety	No. of plants	No. of spikes	Total wt. of grains (g)	Wt. of grains/ spike (g)	1000 grain wt.	No. of grains/ spike
1098–2	10	44	56·0	1·3	35	37
Jyoti	10	42	58·5	1·4	39	36

on the ancillary characters of the line 1098–2 and Jyoti (Table IV) it seems reasonable to conclude that any differences in the grain yield of the two would be accounted for by the thousand grain weight.

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---

1. Chatterjee, S. R., Gassi, Santosh and Abrol, Y. P., *J. Food Sci. and Tech.*, 1974, **11**, 85.
2. Munck, L., Karlsson, K. E., Hageberg, A. and Eggum, B. O., *Science*, 1970, **168**, 985.
3. Chatterjee, S. R., Verma, N. S., Gulati, S. C., Bakshi, J. S. and Abrol, Y. P., *Euphytica*, 1975, **24**, 725.
4. *Official Methods of Analysis*, A.O.A.C., Washington, D.C., 10th Edition, 1965.
5. Chatterjee, S. R. and Abrol, Y. P., Unpublished.
6. — and —, *J. Food Sci. and Tech.*, 1975, Sept.–Oct. issue.
7. Block, R. J. and Mitchell, H. H., *Nutr. Abs. Rev.*, 1946, **16**, 249.
8. F.A.O. Energy and Protein Requirement, *Report of Joint FAO/WHO*, Rome, 1973.
9. Munck, L., Gustaffson, A. and Dormling, I., *Nuclear Techniques for Seed Protein Improvement*, (Proc. Symposium Vienna, 1972), IAEA, Vienna, 1972, p. 203.
10. Hageberg, A.. Karlsson, K. E. and Munck, L., *Ibid.*, p. 121.

ATROPHY OF NODULAR SYSTEM IN THE VIRUS-AFFECTED GROUNDNUT, *ARACHIS HYPOGAEA* L.

Introduction

CHLOROTIC spot virus which produces chlorotic spot disease in the groundnut plant, *Arachis hypogaea* L.[1], seems to exhibit specific metabolic orientation towards γ-methylene glutamine[2] (γ-MG) the reportedly principal transport agent involved in the translocation of nitrogen from seed reserve protein into the growing parts in the early stage of development of the seedling[3]—the cotyledonotrophic stage[4]—prior to the attainment of independence for nitrogen from cotyledonary sources.

The *modus operandi* of the virus exhibiting specific exploitative orientation towards the principal nitrogen source of the fast growing seedling at the stage of preponderant dependence on cotyledonary nitrogen[3] with presumable inability to utilize extra-cotyledonary sources, seems to lead to a severe shift of balance of nitrogen (" nitrogen dysbalance ") heavily tilted towards the virus[4]. This shift creates a " famine of nitrogen amidst plenty of it " in the virus-affected leaves. The nitrogen famine seemingly leads to a lesion oligogynophoria, a " pathological reduction of the number of fruiting gynophores or pathological retardation of the process of fruiting resulting in smaller number of gynophores reaching the stage of fruiting "[5].

Materials and Methods

A batch of healthy and virus-affected groundnut plants was collected from the fields near Nagari, Andhra Pradesh. While uprooting the plants from the soil, care was taken to obtain the root system of the plant in its entirety with minimal loss of lateral roots and nodules. Incidentally the time of explantation of the groundnut plants for the present investigation coincided with the time of fixation of oligogynophoriac lesion reported elsewhere[5]. The number of nodules on the whole root system of each plant was counted. The nodular numerical data of the two groups of plants was processed statistically[6].

Results and Discussion

The nodular numerical abundance is severely affected by viral infection. A statistically significant ($P < 0·001$) reduction by 66·49% is evident in the nodular number on viral infection (Table I). Nodular numerical reduction under viral infection is a common feature[7–10] in plants.

The numerical reduction of nodules in the present case gains ponderable significance since it closely follows oligogynophoriac lesion. Whereas oligogynophoria leads to a pathological reduction of the number of fruiting gynophores[5], the further restriction of nitrogen imposed by reduction of nodular number may lead

TABLE I

Effect of groundnut chlorotic spot virus-infection on the nodular system of the host, Arachis hypogaea L.

Condition	Nodular number per plant's root system.
Healthy (H)	[a]58·0 ± 18·56 (10)
Virus-affected (VA)	19·43 ± 12·68 (7) [b] − 66·49.

[c]*t*: 4·347; *d.f*: 15; P < 0·001

a : Values are means ± standard deviations (numbers of counts).

b : Per cent change of mean nodular number of virus-affected (VA) root system over healthy (H) root system.

c : Statistical treatment:

t = Calculated students, "*t*" test value.

df = number of degrees of freedom.

p = probability of level of significant difference between compared means.

to poorer increase in the size of the "fruiting gynophores". In fact the "virus-affected fruits and seeds" were observed to be consistently smaller than the "healthy fruits and seeds" although no systematic gravimetric and biometric treatment of the fruits and seeds was carried out. Besides, the virus-affected plants, though apparently healthy after the fade-out of symptoms, showed a general but subtle reduction in size of vegetative parts indicating the relative poverty of nitrogen in these plants.

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---

1. Haragopal, T. and Nayudu, M. V., *Phytopath. Z.*, 1971, **71**, 33.
2. —, *Ph.D. Thesis*, Sri Venkateswara University, Tirupati, 1975.
3. Fowden, L., *Ann. Bot. N.S.*, 1954, **18**, 417.
4. Haragopal, T., Jayaramireddy, A. and Chandrasekharam, V., (in preparation), 1975.
5. —, T. and Chandrasekharam, V., *Curr. Sci.* (In press) 1975. *a*
6. Pillai, S. K. and Sinha, H. C., *Statistical Methods for Biological Workers*, Ramprasad and Sons, Agra-3, 1968.
7. Joshi, H. U. and Carr, A. J. H., *J. Gen. Microbiol*. 1967, **49**, 385.
8. Tu, J. C., Ford, R. E. and Quiniones, S. S., *Phytopathology*, 1970, **60**, 518.
9. ———, ———E and Grau, C. R., *Phytopathology*, 1970, **60**, 1653.
10. Rajagopalan, N. and Raju, P. N., *Phytopath. Z.*, 1972, **73**, 285.

EFFECT OF INOCULATION WITH *RHIZOBIUM* AND *AZOTOBACTER* ON NODULATION, GROWTH AND YIELD OF SOYBEAN

IN nature, legumes are bestowed with the advantage of deriving and utilising atmospheric nitrogen in symbiotic association with root nodule bacteria, *viz.*, *Rhizobium*. Several workers[1-3], reported that soybean (*Glycine max.* L.) gave increased yields on inoculation with an efficient culture of *Rhizobium japonicum*. There are reports that *Azotobacter* application exerts a beneficial effect on crop growth which cannot be attributed to an enrichment of the medium with bound nitrogen but to the production of biologically active compounds like indolyl-3-acetic acid, gibberellins which stimulate proliferation of plant seeds and acelerate their growth[4,5]. But in soybean, no studies have been made to see whether the beneficial effect of seed treatment with *Rhizobium* could be further enhanced by soil inoculation with *Azotobacter*. The idea behind this thinking is that if *Azotobacter*, through the production of auxins and gibberellins could stimulate the root elongation and growth, it is creating more area for *Rhizobium* to infect the root system, at as many sites as possible and thereby bring about more nodulation and nitrogen fixation. The present investigation was therefore undertaken to study the effect of soil inoculation with *Azotobacter* on nodulation, growth and bean yield of soybean inoculated with *Rhizobium*.

The field experiment was conducted in a red sandy loam on the experimental farm attached to Main Research Station, University of Agricultural Sciences, Hebbal. A peat-based composite culture of *R. japonicum* was used to treat the soybean seeds variety, Davis. Peat-based culture of *A. chroococcum* (cell count 25×10^8 cells/gm) was applied to soil at the rate of 30 gm per plot. Sufficient farm yard manure (20 tons/ha) was applied uniformly to all the plots before sowing. This is to provide optimum conditions for the growth of *Azotobacter*. The seed was sown in plots of 2·1 M × 2·0 M during September 1974. P_2O_5 as superphosphate and K_2O as muriate of potash were applied as basal dressing at 100 kg and 40 kg. per hectare respectively. The design of

TABLE I

Effect of Rhizobium *and* Azotobacter *inoculation on Soybean*

Treatment	No. of nodules/ plant		Dry wt. of nodules/ plant (mg)		Dry wt. of plant top (gm)		Bean yield/ha. quintals	Percent increase in bean yield over control
	30 days	60 days	30 days	60 days	30 days	60 days		
Control (Uninoculated)	2·3 (1·26)	2·6 (1·55)	4·0 (1·47)	52·0	1·19	2·31	6·37	
Rhizobium only	20·1 (4·49)	47·5 (6·81)	135·0 (11·54)	801·0	1·25	7·32	20·22	217·4
Azotobacter only	8·0 (2·69)	13·4 (3·63)	7·5 (2·56)	325·0	1·27	2·71	12·14	90·6
Rhizobium *Azotobacter*	20·6 (4·55)	44·1 (6·53)	148·0 (11·78)	838·0	1·36	6·16	21·11	231·2
CD at 1 percent	1·48	2·59	2·79	281·97	N.S.	2·11	6·82	
CD at 5 percent							4·86	
SE							3·53	
CV							23·4%	

The values in brackets are transformed values using $\sqrt{x+0\cdot5}$.

the experiment was RBD with five replications of each treatment. The treatment effect was evaluated in terms of nodulation, growth at 30 and 60 days after sowing and final bean yield at harvest.

From the results presented in Table I it can be seen that very few nodules were observed in uninoculated and *Azotobacter* treatments. This can be attributed to the native rhizobia present in the soil. Highest number of nodules was observed in *Rhizobium* treated plants at both 30 days and 60 days of plant growth. Dry weight of the nodules per plant was more or less same in *Rhizobium* alone and *Rhizobium* plus *Azotobacter* treatments. Dry weight of plant tops in various treatments was not significantly different from that of uninoculated plants at 30 days of plant growth. On 60th day, soil inoculation with *Azotobacter* did not show any significant increase in plant weight but seed inoculation with *Rhizobium* resulted in significant increase compared to uninoculated plants and there is no difference between *Rhizobium* inoculation alone and *Rhizobium* plus *Azotobacter* inoculation. Data on bean yield showed that soil inoculation with *Azotobacter* alone yielded 12·14 q/ha. Though the increase in yield from *Azotobacter* treated plots is 90·6% compared to uninoculated control, the plants from *Azotobacter* treated plots showed nitrogen deficiency symptoms during crop growth. This means that the increase in yield may not be due to nitrogen fixation but due to the production of biologically active compounds which stimulated crop growth in the initial stages. Seed inoculation with *Rhizobium* increased the yield by 217·4% and this is not significantly different from the yield obtained with *Rhizobium* plus *Azotobacter* treatment.

It can be concluded from the above findings that in soybean seed inoculation with *Rhizobium* combined with *Azotobacter* inoculation of soil is as good as seed inoculation with an efficient culture of *Rhizobium* in obtaining good nodulation, nitrogen fixation and bean yield and *Azotobacter* inoculation did not contribute either to nodulation or nitrogen fixation efficiency by *Rhizobium*. However, the plants do seem to derive some benefit from *Azotobacter* inoculation alone when compared with the uninoculated control.

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---

1. Agati, J. A. and Garcia, E. H., *Phillip, J. Agric.*, 1940, 11, 271.
2. Erdman, L. W. and Wilkins, F. S., *Iowa Agric. Expt. Stn.* (*Res. Bull.*), 1928, 144, 1.
3. Kumar Rao, J. V. D. K. and Patil, R. B., *Andhra Agirc. J.*, 1974, 21 (In press).
4. Mishustin, E. N. and Shilinkova, V. K., *Biological Fixation of Atmospheric Nitrogen*, MacMillan, London, 1971.
5. Brown, M. E., *Ann. Rev. Phytopath.*, 1974, 12, 181.

EFFECTS OF SOME SPECIFIC INHIBITORS ON THE GERMINATION OF SPORES OF THE FUNGUS *TRAMETES BADIA* (BERK.) COOKE.

THE effects of gibberellic acid on the germination of fungal spores has been well studied[1-4]. The present study attempts to reveal the effect of some specific inhibitors on the germination behaviour of fungal spore, on which much work has not been reported.

Spores of *Trametes badia* were deposited on agar plate. Loopfuls of spores were taken in a test tube containing distilled water. One ml of 50 ppm aqueous solutions of refamycin, 5-fluorodeoxyuridine (FUDR), [2-chloroethyl]-trimethyl-ammonium chloride (CCC) and cycloheximide was poured in petridishes each containing equal amounts of malt-agar media (2·5%) just before solidification. Counting of spores was made in haemocytometer of 0·1 mm depth and the number was calculated in 1 ml of stock solution and was diluted upto 50 spores/ml. One ml of the diluted spore suspension was poured on each of the malt-agar plate containing the respective chemicals and was allowed to stand for a few minutes, water was then decanted off upto the last drop. Control plate was kept side by side. All the petridishes were incubated at 26° C. For studying germination behaviour, each petridish was observed under microscope at 24 hours' interval. At each observation spores from 10 microscopic fields of each of the replicated plates were counted from which the mean was derived. Final result was obtained when 100% germination was achieved in one or the other petridish relative to the rest. Experiments were performed at least twice and gave reproducible results. The whole experiment was performed under sterilized conditions.

Table I shows that after 120 hours 100% spore germination occurred in the control plate, at the same time 34, 42, 86 and 90% inhibition of spore germination was obtained in the plates treated with the chemicals refamycin, FUDR, CCC and cycloheximide respectively.

TABLE I

Percentage of germination of fungal spores treated with some specific inhibitors

Treatments	% of germinated spores				
	24 hours	48 hours	72 hours	96 hours	120 hours
Control	0	72	84	97	100
Refamycin	0	58	62	66	66
FUDR	0	48	51	58	58
CCC	0	6	9	14	14
Cycloheximide	0	3	7	10	10

Contrary to previous findings[2-4] that GA inhibits germination, reports are also available[1] that low concentrations of GA promote germination of fungal spore. CCC is known to be antigibberellic in action[5]. GA has been reported to inhibit the dehydrogenase activity[6] and to interfere with carbohydrate metabolism[7]. The inhibition of spore germination by CCC may therefore be assumed through CCC endogenous GA interaction, though the inhibition of enzymes cannot be ruled out. Cycloheximide has been reported to inhibit enzyme and nucleic acid synthesis in higher plants[8] and in this experiment this chemical has exhibited 90% spore inhibition. Refamycin is an inhibitor of *m*-RNA synthesis and probably acted through the inhibition of RNA-polymerase necessary for the synthesis of *m*-RNA. FUDR has been proved to inhibit DNA synthesis. This chemical might, therefore, decrease the availability of DNA template for the synthesis of *m*-RNA, thereby inhibiting the synthesis of enzymes necessary for the germination of spores. Considering that fungal spore germination is a physiological process it can be concluded that the whole process is enzyme-controlled. Also considering from enzymological point of view, it appears, however, that the degrees of inhibition of spore germination is dependent on the degree of inhibition of enzyme synthesis necessary for the germination by the specific inhibitors.

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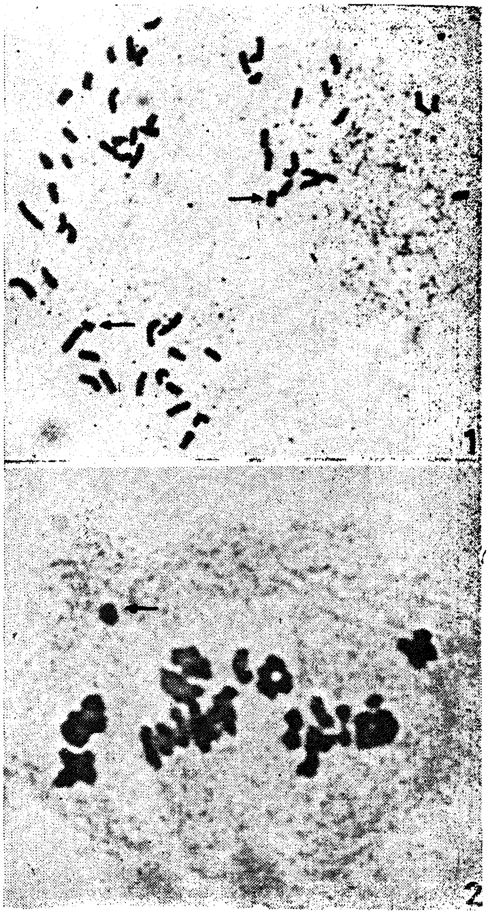
1. Saxena, B. B. L. and Sehra, K. B., *Indian J. exp. Biol.*, 1966, 4, 40.
2. Terui, M. and Kagawa, H., *Chem. Abstr.*, 1962, 56, 726 *g*.
3. Manninger, E., *Naturwissenschaften*, 1961, 41, 508.
4. Teefel, K. and Behnke, U., *Ernahrungsforschung*, 1962, 7, 132.
5. Tolbert, N. E., *Pl. Physiol.*, 1960, 35, 380.
6. Isenberg, F. M. R., Jensen, C. O. and Odland, M. L., *Science*, 1954, 120, 464.
7. Naylor, A. W., *Arch. Biochem. Biophys.*, 1951, 33, 340.
8. Kerridge, D., *J. Gen. Microbiol.*, 1958, 19, 497.

SUPERNUMERARY CHROMOSOMES IN *PENNISETUM SQUAMULATUM* FRESEN.

THE present paper deals with preliminary cytological investigations on *Pennisetum squamulatum* Fresen., a member of Poaceae. It is an autohexaploid ($2n = 54$), an exotic grass introduced from Kenya, Africa. Raman *et al.*[6] reported the occurrence of 25 bivalents and one quadrivalent in this taxon. However various degrees of chromosomal associations during meiotic cycles have been noticed[4]. This note reports the occurrence of supernumerary chromosomes for the first time in this taxon. Material for the present study was collected from Coimbatore.

Distribution of the chromosomes during anaphase I of meiosis shows essentially normal type of disjunction. About 30–40% of the pollen mother cells always show an unequal distribution of 26 and 28 chromosomes. Sometimes 9–11 univalents were observed as laggards of which at least 2–4 chromosomes are eliminated as micronuclei during the formation of pollen tetrads. Pollen sterility was found to be 56% which is indicative of the disturbances occurring in germ cells. While scoring for the different types of associations during meiosis, occasionally in about 50–60% of the PMC's precociously moving one extra bivalent which is globular is being observed along with the regular complement during metaphase (Fig. 2). About 80–90% somatic plates have always two extra chromosomes which are highly heterochromatic (Fig. 1). In somatic tissues these supernumerary chromosomes were either acrocentric or nearly sub-metacentric and remained rod-like. Longley and Randolph were the first to make a clear distinction between the supernumerary chromosomes and the normal complement in plants (cited in Jones)[3]. The incidence of supernumerary chromosomes among the species of Poaceae is large and variable according to Jones. Reports regarding such chromosomes in the tribe Paniceae are not common. In two diploid species of *Pennisetum*, *P. typhoides* and *P. orientale*, supernumerary chromosomes have been recorded by Pantulu[5] and Jauhar and Singh[2] respectively. Supernumerary chromosomes are believed to be formed either by fragmentation of certain chromosomes of the normal complement or by non-disjunction[1]. Of the four cells present in the pollen tetrad only 1 or 2 had micronuclei ; the number of micronuclei varied from 1–2. The supernumerary chromosomes seem to be eliminated during meiotic cycles. Moreover cyto-embryological studies also indicate that this grass is a facultative apomict, the mechanism of reproduction being aposporic. Detailed study of this aspect is in progress.



FIGS. 1–2. Fig. 1. Somatic chromosomes of root tips. $2n = 54 + 2$, × 2,800. Fig. 2. Meiosis—Metaphasic view, × 2,800. Arrows indicate supernumerary chromosomes.

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---

1. Brown, W. V., *Text-book of Cytogenetics*, Mosby and Co., 1972.
*2. Jauhar, P. and Singh, *Genet. Iber.*, 1970, 22, 53.
3. Jones, R. N., *Inter. Rev. Cytol.*, 1975, 40, 1.
4. Narayan, K. N. and Nagabhushana Rao Sindhe, A., *Proc. 57th Ind. Sci. Cong. Abstr.*, 1970, **84**, 285.
5. Pantulu, J. V., *Curr. Sci.*, 1960, **29**, 28.
6. Raman, V. S., Chandrasekharan, P and Krishnaswamy, D., *Ibid.*, 1959, **28**, 127.

* Not seen in original.

SONERILA PEDUNCULOSA THW. (MELASTOMATACEAE)—A NEW RECORD FOR INDIA

A POPULATION of *Sonerila* Roxb. was collected from Charmadi Ghats, Chikmagalur District, Karnataka State, during the monsoon of 1972. It was identified as *S. pedunculosa* Thw.

While Trimen in *Fl. Ceylon* 2 : 208. 1893 treats *S. pedunculosa* as endemic to Ceylon, Clarke in Hk. f. *et al., Fl. Brit. India* 2 : 531. 1879 records it also from Quilon, Travancore (India) on the basis of a specimen *Wight* 1111. Our efforts (personal communication) revealed that *Wight* 1111. deposited at K should be identified as *S. rheedii* sensu Wt. and Arn. and not *S. pedunculosa* as stated by Clarke (*l.c.*). Further, our specimen *Yoganarasimhan* 1331 was identified as *S. pedunculosa* at Peradeniya herbarium, Sri Lanka and at CAL. At Kew (K) it was identified as *S. rheedii* on the basis of *Wight* 1111 which itself represents a mixture, at least on the Kew sheet (personal communication). A comparison of *Yoganarasimhan* 1331 with *S. rheedii* at MH showed that they are different. Further, Dr. M. P. Nayar who has carried out extensive revision work on the S. E. Asian Melastomataceae during his tenure of work at Kew on re-examination of *Yoganarasimhan* 1331 confirmed it as *S. pedunculosa* only (personal communication). Also, *S. pedunculosa* can easily be distinguished[1] from *S. rheedii* in the prostrate and creeping habit and capsules provided with thick valves instead of the erect habit and capsules provided with thin valves ; the former characters were characteristically observed in the population of *S. pedunculosa* (*Yoganarasimhan* 1331).

Hence, giving more credence to the identification at Peradeniya (type locality) supported by the expert opinion of Nayar, and the fact that it does not match with *S. rheedii, Yoganarasimhan* 1331 is identified as *S. pedunculosa,* which incidentally forms a new record for India. A short description of the taxon along with a plate of herbarium specimen follows.

Sonerila pedunculosa Thw. *Enum. Pl. Zeyl.* 109. 1859 ; Cl. in Hk. f. *et al., Fl. Brit. India* 2 : 531. 1879, *p.p.. quoad. typum* excl. syn. *et* sp. *Wight* 1111 ; Trimen, *Fl. Ceylon* 2 : 208. 1893.

Procumbent herbs ; stems rooting at nodes, glabrous, succulent, internodes long. Leaves up to 6 × 3 cm, ovate-lanceate, subobtuse or rounded at apex, finely crenate serrate, with numerous scattered hairs above, nearly glabrous beneath, flaccid, penninerved, base rounded ; petioles 1–2 cm long. Flowers pinkish, small, in racemes, at ends of long, erect, slightly glandular, pilose peduncles, terminating in short apical bracts ; calyx with scattered, glandular hairs ; anthers stout, oblong ; dehiscence poricidal. Capsules small, turbinate, smooth, faintly ridged, valves thick ; seeds minute, numerous (Fig. 1).

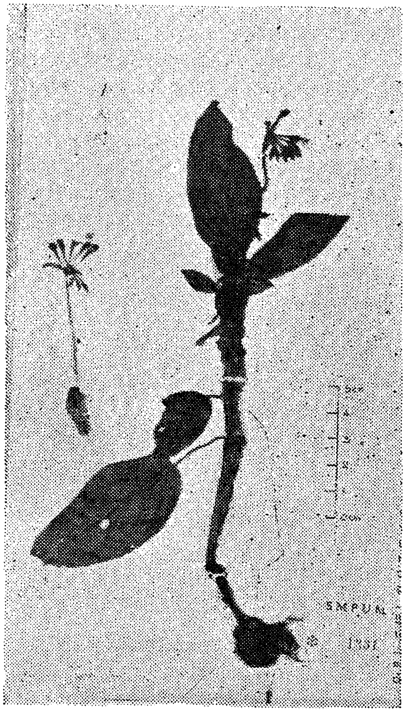

FIG. 1

Herbarium specimen examined : *Yoganarasimhan* 1331, near Annappa falls, Charmadi Ghats ; in moist rocky slopes along streams, *ca* 1000 m, fairly common, flowering and fruiting August, deposited at Herbarium of Regional Research Centre, Bangalore. The present finding represents an extension of its distribution in South India.

We are thankful to the Directors of the herbaria at Kew, Peradeniya, Calcutta, and the Regional Botanist, Coimbatore, for help rendered ; to Drs. M. P. Nayar and A. N. Henry, for their valuable opinion ; the Director, CCRIMH and Officer-in-Charge, Regional Research Centre, Bangalore, for facilities ; one of us (K. S.) is thankful to the University Grants Commission, for financial assistance.

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Mysore 570 006, *March* 25, 1976.

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1. Nayar, M. P., *J. Indian Bot. Soc.*, 1969, 48, 321.

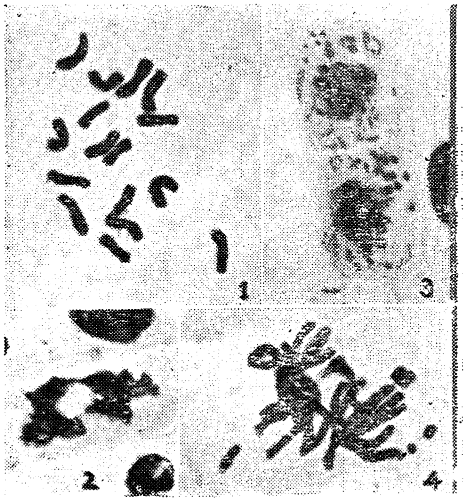
THE EFFECTS OF THE GLYCOSIDE "MORINDINE" ON MITOTIC CHROMOSOMES

In view of the wide distribution in plants and in possessing physiological action most of the glycosides like digoxin and digitoxin (cardiac glycosides) have a therapeutic application in medicine. The antimitotic actions of Nerifolin, Thevetin[1], Digitonin[2, 3] and Saponin fractions[4] have been described. The water extracts of *Nerium oleander* are known to possess anticancer properties[5] and is used for heart malfunctions. The antimitotic activity of the glycosides present in the extract has been demonstrated[6, 7]. Still there are several glycosides whose cytological effects have not been studied. As our knowledge on the effects of glycosides on mitosis is fragmentary the present studies have been undertaken.

The genus *Morinda* (Rubiaceae) is economically important in that the various parts of the plant are used in dyeing textiles and in medicine[8]. The glycoside "Morindine" (6-Primeverose of Morindone $C_{26}H_{28}O_{14}$) has been extracted and purified from the root bark of *M. tinctoria* var. *tomentosa* Hook. f. (Rao and Reddy, unpublished).

Root tips from the bulbs of *Allium cepa* were grown in 0·05% aqueous solution of "Morindine" and fixed in one series after 4, 8 and 24 hour treatments, while in another the bulbs treated for 24 hours were allowed to recover in distilled water for 24 and 48 hours. Controls were grown under identical conditions. The cytological preparations were made by the method as described elsewhere[9].

As compared to the control the mitotic index (M.I.) showed a reduction with the period of treatment thus denoting the antimitotic activity. The recovery experiments, however, showed a return to the level of the control from 24 to 48 hours indicating a slow reversal from the damage (Table I). The chief effect of the chemical was one of partial C-mitotic effect which increased with the period of treatment upto 8 hours (Fig. 1), but in 24 hour treatment the number of C- metaphases had shown a reduction. There was also a concomitant reduction in the M.I. (Table I).



FIGS. 1–4. Haematoxylin squashes of *Allium cepa* after treatment with Morindine, × *ca.* 1100.

The other aberrations in the order of severity were stickiness, sticky bridges (Fig. 2), cells with dispersed chromatin (Fig. 3) and also chromosome breakages (Fig. 4) only after 24 hour treatment. The rare instances of cells with dispersed chromatin are indicative of nuclear degeneration[10]. Their absence after recovery shows that such pycnotic cells have degenerated completely. These studies reveal that like other glyco-

TABLE I

Treatment time (in hrs.)	Frequency of aberrations*				
	Miotic index	C-metaphases	Stickiness	Bridges	Breakages
Control	5·81	..	..	..	..
4	5·60	18	9	..	..
8	5·34	30	14	4	..
24	3·80	22	12	..	10
24 hr. treatment					
24 hr. recovery	4·60	16	2	..	..
48 hr. recovery	5·78	..	..	..	..

* Based on 100 metaphases and anaphases.

sides, Morindine has an antimitotic activity. The partial damage caused to the genetic system by it is a slow but reversible process.

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1. Vcnkateswarlu, J. and Srinivasan, K., *Mem. Ind. Bot. Soc.*, 1960, **3**, 78.
2. Olah, L. V., *Proc. Int. Cong. Genetics*, *11th, The Hague*, 1963, **1**, 102.
3. —, *Bull. Torrey Bot. Club*, 1965, **92**, 197.
4. Oswiecimska, M., Sendra, J. and Janeczko, Z., *Polish. Jour. Pharmacol and Pharmacy*, 1975, **27** (3), 349.
5. Taylor, A., McKenna, G. F. and Burlage, H. M., *Texas, Reports on Biology and Medicine*, 1956, **14**, 538.
6. Tarkowska, J. A., *Acta Soc. Botani. Poloniae*, 1971, **40**, 623.
7. —, *Hereditas*, 1971, **67**, 205.
8. Bhatnagar, S. S., Chopra, R. N., Baini Prasad, Ghosh, J. C., Quereshi, M., Saha, M. N., Lala Sri Ram and Manjunath, B. L., *The Wealth of India. A Dictionary of Raw Materials and Industral Products*, Vol. VI, L-M, C.S.I.R., New Delhi, 1962, p. 423.
9. Subramanyam, S. and Subramaniam, M. K., *Proc. Indian Acad. Sci.*, 1970, **72B**, 115.
10. Makinen, Y., *Ann. Bot. Soc.* " *Vanamo* ", 1963, **34** (7), 1.

' CHEMICAL DIMORPHISM ' OF POLLIEN GRAINS OF RUBIACEAE

Dimorphism of pollen grains for various palynological characters such as pollen size, size of ora, aperture number, exine thickness and ornamentation has been reported in a number of angiosperm families including Rubiaceae[1–4]. The pollen grains of a few South Indian species of Rubiaceae have been found to exhibit another dimorphic feature characterised by colour difference of the exine after acetolysis. This note is a preliminary report of this phenomenon observed in nine species.

Polliniferous materials were procured from live plants, and pollen preparations made by acetolysis method[5]. After acetolysis, the pollen grains of these species showed noticeable difference in their colour, some of them being dark brownish and others clearly lighter, and both types occurred in appreciable frequencies (Table I). This dimorphic feature was consistently observed in populations of these species assembled from various localities.

Table I

Frequency of dark coloured and light coloured pollen grains

Species	Dark coloured grains %	Light coloured grains %
Oldenlandia diffusa Roxb.	20·0	80·0
Neanotis monosperma Wallex wt and Am	46·5	53·5
Guettarda speciosa Linn.	27·3	72·7
Knoxia sumatrensts (Rets) DC	26·4	73·6
Cantium rheedii DC	17·6	82·4
C. neilgherrensis Bedd	55·4	44·5
Morinda citrifolia Linn.	43·6	56·4
Psychotria globicephala Gamble	24·3	75·7
Borreria eradii Ravi	35·4	64·6

It appears that the difference in colour is due to deep staining of the endoexine in some grains, and this was particularly evident in two of the species, *Knoxia sumatrensis* and *Canthium neilgherrensis*. The phenomenon of colour difference noticed here may be taken as evidence of ' chemical dimorphism ', because the difference is noticeable only after the chemical treatment. The pollen mass consists of pollen strains of different physiological potential, reflected in morphological differences, and the present finding may be another evidence to substantiate such a contention.

We are indebted to Prof. C. A. Ninan, for encouragement and facilities, and to Dr. P. K. K. Nair, National Botanic Gardens, Lucknow, for critical examination. One of us (O.P.) is thankful to the CSIR, Government of India for the award of a Research Fellowship.

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November 28, 1975.

---

1. Baker, H. G., *Evolution*, 1956, **10**, 23.
2. Lewis, W. H., *Grana Palynol.*, 1964, **5**, 330.
3. Bir Bahadur, *Rhodora*, 1964, **66**, 56.
4. Mathew, P. M. and Omana Philip, *J. Palynol.*, **12** (in press).
5. Nair, P. K. K., *J. Sci. Ind. Res.*, 1960, **19**, 26.

SHORT SCIENTIFIC NOTES

Leaf Spot Disease of Rubber Caused by *Curvularia pallescens* Boedijn

During months of June–July, minute brownish red spots with yellow halo were noticed on leaves of *Hevea brasiliensis* in the budwood nursery. From these spots *Curvularia pallescens* Boedijn was isolated.

The fungus grows well on potato dextrose agar and oats agar. The culture was slightly greenish in colour in the beginning but later on developed a black velvety, glistening appearance with its conidia and conidiophores.

Artificial inoculation on rubber seedlings in the field and on cut shoots in the laboratory produced high percentage of infection. Young flushes sprayed with a conidial suspension from 7-day-old culture of the pathogen produced brownish red spots within 24 hours. The spots are slightly raised at the lower surface of the leaf. Shrivelling of the leaves was noticed due to the infection at the copper brown stage. *Curvularia pallescens* Boedijn was isolated from lesions produced by artificial inoculation.

The author expresses her sincere thanks to Shri. P. N. Radhakrishna Pillai, Deputy Director and the Director, Rubber Research Institute of India, for their encouragement and to CMI, Kew, England, for kindly identifying the pathogen.

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An Analysis of Step-Drawdown Test Data—A Simplified Solution

The analysis of step-drawdown test for determining well loss constant and aquifer loss constant from Jacob's equation (1946) has been simplified by merely solving the drawdown equations of the steps at a time, like simultaneous equations. The step-drawdown test data from Betul area in Madhya Pradesh conducted by the Evangelic Luther Church Water Development Project have been taken to elucidate this simplified method. The test data were analysed using the graphical method of Bruin and Hudson (1955) in the above project. The value of B, *i.e.*, the aquifer loss constant, and C, the well loss constant, by the graphical method were determined to be 0·1225 and 0·00072 respectively.

The values from the test data were introduced in Jacob's equation. The simultaneous equations for the two steps are as follows :

$$0{\cdot}14 = B + C \times 25 \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots \text{I}$$

and $$0{\cdot}146 = B + C \times 33{\cdot}33 \quad \ldots\ldots\ldots\ldots\ldots\ldots \text{II}$$

After solving the simultaneous equations I and II, the value of C comes out to be 0·00072. Introducing the value of C in equation I the value of B comes out to be 0·122. Similarly taking into consideration the third and fourth steps, the equations may be written as follows :

$$0{\cdot}15 = B + C \times 41{\cdot}66 \quad \ldots\ldots\ldots\ldots\ldots\ldots \text{III}$$

and $$0{\cdot}164 = B + C \times 59{\cdot}00 \ldots\ldots\ldots\ldots \quad \text{IV.}$$

After solving III and IV the value of B and C are 0·1162 and 0·00081 respectively.

From the above it is seen that the values of B and C calculated by the graphical method and from the proposed simplified solution are comparable. Hence this proposed method of calculating aquifer loss constant and well loss constant may be used conveniently.

Central Ground Water Board, P. G. ADYALKAR.
Nagpur, *November* 27, 1975. V. V. S. MANI.
S. K. SHARMA.

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1. Bruin, J. and Hudson, H. E. (Jr.), "Selected method for pumping test analysis," *Ill. Sta. Wat. Surv. Rep.*, 1965, No. 25.
2. Jacob, C. E., "Drawdown test to determine effective radius of artesian well," *Proc. Am. Soc. Civil Engrs.*, 1946, **5**, 72.

A New Disease of Sword Bean

The authors have come across an interesting disease of sword bean (*Canavalia ensiformis* DC). The disease initially shows dark-brown to black spots on the fruit, which later on coalesce to form irregular lesions. In the advanced stage not only the lesions assume bigger size but also small round black bodies make their appearance. Isolations from the diseased portions revealed it to be *Coleophoma empetri* (Rostr.) Patrak in culture (IMI 190741).

The fungus was claimed as pathogen only after satisfying the Koch's postulates. The artificial inoculations were successful when the fruits were injured with the help of sterile scalpel. Perusal of the literature[1] revealed that it has not been reported either from India or elsewhere. This forms the new genus record for India.

Thanks are due to Dr. Johnston and Dr. Sutton of Commonwealth Mycological Institute, Kew, England, for the identification of the fungus and to Prof. U. B. S. Swami for his kind encouragement and laboratory facilities.

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December 3, 1975.

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1. Sam Raj, J. and Jose, P. C., *Sci. and Cult.*, 1969, **35**, 623.

A New Leaf Blight of *Peristrophe bicalyculata*

Peristrophe bicalyculata L. grows abundantly as a weed in gardens, wastelands, grasslands and along the sides of footpath and roads. A severe leaf blight resulting in defoliation was observed on this plant in Meerut during September–October, 1975. The chief characteristic symptoms on the leaf are brownish, depressed, necrotic areas, surrounded by reddish to dark brown, slightly raised margins. The disease appears as small, round, brownish spots which become darker as they enlarge. Such spots sooner or later coalesce with each other forming large necrotic areas, chiefly along the margins of leaves. These lesions sooner or later are shed from leaves, leaving behind distinct holes. Severely infected leaves tend to roll up from the margins, get shrivelled and finally get detached from the plant.

Microscopic examination of preparations from diseased leaves and culture prepared from such leaves revealed the presence of a fungus, which was identified as *Colletotrichum crossandrae* Patel, Kamat and Pande. Pathogenicity tests were made by making cross inoculations on the present host and on three other plants, *Crossandra infundibuliformis*, *Ruellia tuberosa*, and *Barlaria* sp. of the host family. Successful infection, with symptoms characteristic to those observed in the field, developed only on the host plant indicating the host specificity of the present fungus. Cross inoculations of leaves of the above-said four plants with the isolate from *Crossandra infundibuliformis*, showed the development of characteristic symptoms only on *C. infundibuliformis*.

Although, minor insignificant differences in size of aceruvuli, setae and shape and size of conidia, in its morphological characters the present fungus resembled *Colletotrichum crossandrae*, reported earlier from India on *Crossandra infundibuliformis*[6]. Athongh, shape and size of conidia, presence or absence of sclerotia and chlamydospores, shape and size of appressoria and acervuli, host range[12, 13] and pathogenicity and cross inoculation tests[3, 4, 5, 13] have been used in the past as the criteria in delineation of species in the genus *Colletotrichum*, morphologic features and host specificity of the present fungus, as reported herein, seem hardly of a magnitude for a justified differentiation of a new species in the genus. More or less similar views about morphologic characters[7, 9, 11] and host specificity[1, 8–10] as criteria of less value for delineation of species in this genus have also been expressed earlier. Since the fungus reported herein is host-specific, and when compared with *Colletotrichum crossandrae* shows insignificant variations only in respect of some of the morphologic features, it seems likely that the present fungus is an ecological variant of *C. crossandrae*. It has already been shown earlier that dimensions of acervuli, setae, etc., fluctuate in the same species of *Colletotrichum* on different hosts[2] and being influenced by the environment and the substratum[8].

The herbarium specimen of the type material and culture (C.H. MCM, 205) are deposited in the cryptogamic herbarium, School of Plant Morphology, Meerut College, Meerut.

Department of Botany, P. D. SHARMA.
Meerut College, Meerut, D. K. SAINGER.
October 17, 1975
(Revised *May* 24, 1976).

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*1. Berger, O. F., *J. Agric. Res.*, 1921, **20**, 725.
2. Butler, E. J., *Fungi and Diseases in Plants*, Thacker Spink and Co., Bombay, 1918.
3. Dastur, J. F., *Indian J. Agric. Sci.*, 1934, **4**, 100.
*4. Hemmi, T. and Kurata, S., *J. Soc. Trop. Agric. Taiwan*, 1935, **6**, 573.
*5. Lee, H. A., *Philip. J. Sci.*, 1924, **22**, 603.
6. Patel, M. K., Kamat, M. N. and Pande, C. B., *Indian Phytopath.*, 1952, **5**, 130.
7. Quinione, S. S., *Thesis Associateship of I.A.R.I.*, New Delhi, 1958.
8. Ramakrishnan, T. S., *Proc. Ind. Acad. Sci.*, 1941, **13 B**, 60.
9. —, *Ibid*, 1947, **15 B**, 15.
10. Singh, R. D., Prasad, N. and Mathur, R. L., *Indian Phytopath.*, 1966, **19**, 65.
11. Small, W., *Trans. Brit. mycol. Soc.*, 1926, **11**, 112.
12. von Arx, J. A., *Phytopath. Z.*, 1957 *a*, **29**, 413.
13. —, *Tijdschr Plziekt*, 1957 *b*, **63**, 171.

* Original not seen.

REVIEWS AND NOTICES OF BOOKS

Taxonomy of Indian Mosses. By R. S. Chopra. (Botanical Monograph No. 10, Publications and Information Directorate, C.S.I.R., New Delhi), 1975. Pp. XL + 631. 16 cm × 24 cm. Price Rs. 96·00 (Inland), $ 38.00, or £ 15.00 (Foreign).

The moss flora of the subcontinent of India is quite rich and in spite of this, these fascinating groups of plants did not attract sufficient attention from adequate number of research workers. Hence, it is most opportune that the *Taxonomy of Indian Mosses* by R. S. Chopra, who is well known for his authoritative knowledge on these plants, has been published by the Council of Scientific and Industrial Research. This treatise on mosses is a result of Dr. Chopra's careful and intensive studies extended over a period of more than thirty years.

After an editor's note and a brief preface, and a conspectus of the various classes and subclasses, they are followed by an elaborate taxonomic account of the Musci. In all, a total number of nearly 2,000 species of mosses belonging to 329 genera under 56 families are treated in a very detailed manner. While dealing with orders, families and genera the classification of Brotherus (1924–25) has been followed with the exception that Fissidentales follows the Dicranales. In the case of Pottiaceae Chen's (1941, 1941 *a*) classification has been adopted in general.

The systematic account of Musci (Muscophytina) starts with a key to the classes and subclasses. In each order and subclass, the general characters of the order is followed by the family and then the key to the genera and species. Author citations are indicated at appropriate places. The entire work is profusely illustrated with as many as 122 carefully written line drawings. These are particularly helpful because they relate to the exomorphic and endomorphic characters of each plant, thus serving in their easy identification; further, the drawings greatly enhance the value and usefulness of this work. The descriptions for each taxon are very well written and throughout a uniformity is maintained.

An extensive citation of references is another redeeming feature of this work. A carefully selected glossary of technical terms is provided for easy understanding. The index at the end is helpful for ready reference.

The chief editor has mentioned that the *Taxonomy of the Indian Mosses* is the last and most voluminous of the monographs of the series and this is rightly so because this excellent monograph will end as the largest monograph in this series.

Dr. Chopra has to be congratulated for writing this taxonomic treatise on Indian Mosses and the Publication and Information Directorate of the C.S.I.R. for bringing out this botanical monograph in such a neat and elegant manner maintaining as usual their technical excellence. This work must find a place in all Universities and Research Institutions and to students of Musci in particular, it will serve as a valuable reference work in the years to come.

K. SUBRAMANYAM.

Introduction to Computers and Programming. By K. Srinivasa Rao. (S. Viswanathan, Printers and Publishers, Pvt. Ltd., Madras), 1974. Pp. xiv + 329. Price Rs. 15·00.

This is a good introductory book on Computers and Programming. The book consists of 11 chapters. The first two chapters present the salient features of digital computers. In the III Chapter, the computer's arithmetic is described. Chapters IV to IX deal with FORTRAN language. Chapter X deals with the types of computers, the number of computers and their location in India. Chapter XI gives a brief review of the applications of computers in the field of science, engineering, space research, sports, etc.

Needless to say that there is nothing new except the way of presentation. The book is well written. It is easily understandable and it does not require previous knowledge of computer or programming. However, FORTRAN programmes for numerical integration of differential equations, etc., could have been easily included in the book without any difficulty. On the whole, this book packs a lot of information and it will be valuable to anyone who is interested in the elementary knowledge of the subject.

G. NATH.

5896/76. **Published by S. R. S. Sastry, Manager,** Current Science Association, Bangalore-560006
and Printed at The Bangalore Press, Bangalore-560018.

CONFORMATION OF RANDOM POLYNUCLEOTIDE CHAINS—FREE ROTATIONAL DIMENSIONS

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ABSTRACT

Free rotational dimensions (C_f) of polynucleotide chains, with four different types of rigid sugar residues, have been computed as a function of the linkage angle (τ), using the single virtual bond approach. The computed C_f values for the freely rotating polynucleotides are close to unity, indicating that the above polymer behaves quite similar to freely jointed chains. No significant difference is found in the computed C_f values for a fixed τ, for all the four types of polynucleotides. Small variations in τ do not effect C_f values significantly.

INTRODUCTION

THE stiffness of polymeric chains is generally characterised by $\sigma = \{\langle r^2\rangle_0/\langle r^2\rangle_{0,f}\}^{1/2}$ where $\langle r^2\rangle_0$ is the unperturbed mean square end-to-end distance and $\langle r^2\rangle_{0,f}$ is that calculated from a model assuming free rotation of the monomeric units about the single bonds linking the adjacent units. The free rotational dimensions for polypeptides[1,2] and polysaccharides[3] have been reported by treating the chains comprising N monomeric units as a sequence of N virtual bonds of constant length l_v. Similar methods have been followed in computing the dimensions of hindered polypeptides[1,2,4,5], polysaccharides[6-8] and polynucleotides[9]. Presently, we have attempted to compute the $C_f\ (= \langle r^2\rangle_{0,f}/Nl_v^2)$ values of freely rotating polynucleotides following the single virtual bond scheme[a] by Olson[9]. The polynucleotides have been assumed to comprise of four types of rigid sugar residues, *viz.*, C_3'-endo, C_2'-exo, C_2'-endo and C_3'-exo with a fixed valence angle (τ) at the linking phosphorus atoms. In view of the observed variations of this angle from 100° to 106°[10-14], a systematic study was undertaken to compute the C_f values using the single virtual bond approach for polynucleotides with four different types of rigid sugar residues, as a function of τ. These results may help in understanding the effect of variations in the linkage angle on the free-rotational dimensions and on comparison with the experimental values, may also furnish information about the stiffness of the various polynucleotide chains.

THEORETICAL TREATMENT

A section of the backbone of a polynucleotide chain showing the virtual bonds is shown in Fig 1. The bond lengths and valence bond angles of the chain backbone were taken from the compilation of Arnott[14]. The sugar residues in the different polynucleotides studied in the present work were fixed in C_3'-endo, C_2'-exo, C_2'-endo and C_3'-exo conformations with the rigid nucleotide geometries used by Olson[9]. The recently recommended[16] notation and conventions were followed in describing the chain parameters.

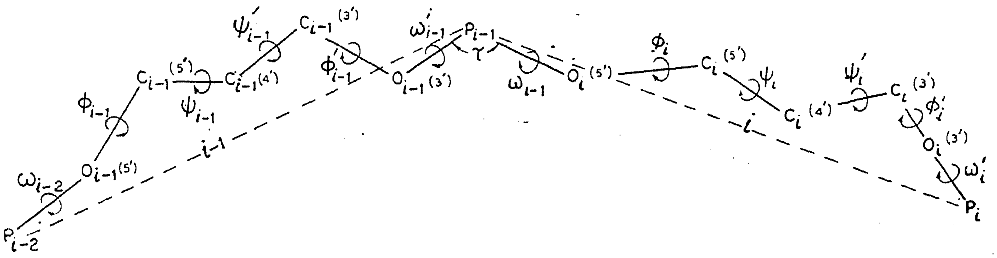

FIG. 1. Section of the backbone of a polynucleotide chain showing the rotational angles and virtual bonds (dashed lines).

The general expression for calculating the chain characteristic ratio of an infinitely large polynucleotide chain was obtained by using the expression[9]

$$\langle r^2\rangle_0/Nl_v^2 = [(E + \langle T\rangle)\,(E - \langle T\rangle)^{-1}]_{22} \qquad (1)$$

---

[a] *Foot Note* '*a*': The rigid nucleotide concept and the connected virtual bond scheme have evolved from the original work of M. Sundaralingam and his associates. (Sundarialingam, M. (1973). In 'Conformation of Biological Molecules and Polymers', *The Jerusalem Symposium on Quant. Chem. Biochem.* Ed. Bergmann and B. Pullman, 5, 417–456, Academic Press, New York; Yathindra, N. and Sundaralingam, M. (1975), In *Structure and Conformation of Nucleic Acids and Protein—Nucleic Acid Interaction*, Ed. M. Sundaralingam and S. T. Rao, 729–748, University Press, Baltimore).

were E is the identity matrix of order 3 and $\langle \mathbf{T} \rangle$ is the statistical mechanical averaged matrix. (However, Olson[9] expresses the characteristic ratio as $\langle r \rangle_0/nl^2$ where n is the total number of chemical bonds in the chain backbone and l^2, the mean square bond length). The subscript 22 denotes the (2, 2) element of the final matrix. The transformation matrix T_i, which transforms a vector presented in the coordinate system associated with the virtual bond i to the coordinate system of i–1 was formed following the procedure of Rao *et al.*[6]

RESULTS AND DISCUSSION

Since in a freely rotating polynucleotide chain all the orientations of the monomer units are equally probable, the characteristic ratios, C_f, of such a chain may be calculated by using equation 1 by giving equal statistical weights to all the conformational states. The C_f values computed for polynucleotides with different types of sugar puckerings for different values of τ are given in Table I. It is seen from Table I that the computed C_f values for the freely rotating polynucleotide chains are close to unity, indicating that the above polymer behaves quite similar to freely jointed chains. This happens because the angle between the virtual bonds varies (though the angle τ is kept constant) when the nucleotide units are rotated about the linking single bonds. Thus the chain formed by the virtual bonds tends to attain the character of a freely jointed chain. No significant difference is found in the computed C_f values for a fixed τ for all the four types of polynucleotides. This indicates that the different rigid geometries for the nucleotides in the polynucleotide chains have little effect on the free rotational dimensions.

TABLE I

Calculated values of the characteristic ratio C_f of freely rotating linear polynucleotide chains.

Type of sugar puckering in the polynucleotide chains	Values of C_f for different values of angle τ				
	100°	101·4°	103°	104·5°	106°
C_3'-endo	1·12	1·13	1·15	1·17	1·18
C_2'-exo	1·13	1·14	1·16	1·18	1·20
C_2'-endo	1·21	1·24	1·27	1·30	1·33
C_3'-exo	1·23	1·26	1·30	1·34	1·38

Table I also shows that there is no significant change in the C_f values of all the polynucleotide chains for different τ values. This again indicates that the free rotational dimensions of polynucleotides do not depend on the valence angle at the linking phosphorus atom. This behaviour is in contrast to that of polypeptides[2] and polysaccharides[3] for which C_f is generally found to increase significantly with increase in τ.

Note added in proof: Since this paper was submitted for publication the author has learnt from Dr. N. Yathindra of the University of Wisconsin that he has obtained similar results in independent calculations but his results are yet unpublished.

1. Brant, D. A., Miller, W. G. and Flory, P. J., *J. Mol. Biol.*, 1967, **23**, 47.
2. Srinivasan, A. R. and Rao, V. S. R., *J. Polym. Sci.*, 1970, A–2, **10**, 693.
3. Yathindra, N. and Rao, V. S. R., *Ibid.*, 1971, A–2, **9**, 1149.
4. Srinivasan, A. R. and Rao, V. S. R., *Pramāṇa* 1975, **4**, 95.
5. Brant, D. A. and Flory, P. J., *J. Amer. Chem. Soc.*, 1965, **87**, 2791.
6. Rao, V. S. R., Yathindra, N. and Sundararajan, P. R., *Biopolymers*, 1969, **8**, 325.
7. Yathindra, N. and Rao, V. S. R., *Ibid.*, 1970, **9**, 783.
8. — and —, *J. Polym. Sci.*, 1972, A–2, **10**, 1369.
9. Olson, W. K., *Macromolecules*, 1975, **8**, 272.
10. Shefter, E., Barlow, M., Sparks, R. A. and Trueblood, K. N., *Acta Cryst.*, 1969, **25 B**, 895.
11. Viswamitra, M. A., Reddy, B. S., Lin, G. H. Y. and Sundaralingam, M., *J. Amer. Chem. Soc.*, 1971, **93**, 4565.
12. Sussman, J. L., Seeman, N. C., Kim, S. H. and Berman, H. M., *J. Mol. Biol.*, 1972, **66**, 403.
13. Rubin, J., Breman, T. and Sundaralingam, M., *Biochemistry*, 1972, **11**, 3112.
14. Hingerty, B., Subramanian, E., Stellman, S. D., Broyde, S. B., Sato, T. and Langridge, R., *Biopolymers*, 1975, **14**, 227.
15. Arnott, S., *Prog. Biophys. Molec. Biol.*, **21**, 1970, 265.
16. "Recommendations of standard conventions and Nomenclature for the Description of the Conformation of Polynucleotide chains" (1973) in *Conformation of Biological Molecules and Polymers*, Bergmann, E. D. and Pullman, B., Eds., Israel Academy of Sciences and Humanities, Jerusalem.

SYNTHESIS OF 2-*o*-AZIDOPHENYLBENZOTHIAZOLES AND THE FUSED HETEROCYCLES DERIVED THEREFROM

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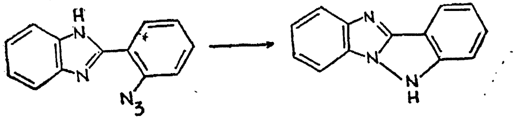
INTRAMOLECULAR insertion reactions of suitably substituted nitrenes have become the basis of a general method of synthesis of various heterocyclic systems[1]. In the majority of these instances, azides are involved as nitrene precursors. Relatively little is known about the intramolecular reactions of nitrene function with a $>C=N-$ component of a heterocyclic system. The only known example of such a heterocyclisation involving 2-*o*-azidoarylazoles is the formation of 6 H-indazolo (2, 3-*a*)-benzimidazole (II) on pyrolysis of 2-*o*-azidophenylbenzimidazole (I)[2].



I II

CHART-I

With a view to studying the formation of the new heterocyclic systems based on this approach, the synthesis of 2-*o*-azidophenylbenzothiazoles and their decomposition under thermal and photochemical conditions have now been undertaken.

A convenient method for the synthesis of 2-*o*-azidophenylbenzothiazole (III, X = H) has been developed in the present investigation, by the condensation of zinc salt of *o*-aminothiophenol with *o*-azidobenzoic acid in the presence of polyphosphate ester. This method has been extended to the synthesis of three other benzothiazoles (Table I). The I.R. spectra of all these compounds showed a strong absorption at 2120–2140 cm^{-1}, characteristic of the azido group[3].

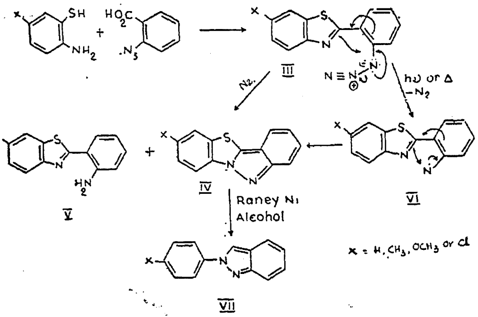


CHART-II

TABLE I

Sl. No.	2-*o*-azidophenylbenzothiazoles (III)			Indazolo (2, 3–*b*) benzothiazoles (IV)		
	X	m.p.	% yield	X	m.p.	% yield
1.	H	133	50	H	143	75–80
2.	CH_3	108	55	CH_3	207	85
3.	CH_3O	158	60	CH_3O	163	90
4.	Cl	124	52	Cl	148	80–85

Cyclisation of 2-*o*-azidophenylbenzothiazole, as a representative case, has been studied in detail, under thermal and photochemical conditions. The exposure of benzene solution of III (X = H) to sunlight or U.V. light for three hours resulted in the formation of a crystalline product (IV), m.p. 143°, in 60% yield. However, the decomposition of the azide, under these conditions, was found to be incomplete. The mass spectrum of the compound (IV) revealed the molecular ion at m/e 224 and showed a strong band at 1650 cm^{-1} in the I.R. spectrum. The absence of the azido function in the I.R. spectrum and the molecular weight of the compound strongly suggest indazolo (2, 3-*b*) benzothiazole structure for the compound (IV). It may be mentioned here that similar I.R. absorption has been reported in the case of pyrido (1, 2-*b*)-indazole[4] and has been attributed to the dipolar canonical form. Pyrolysis of III (X = H) with sand for 45 minutes at 160° yielded, in addition to IV (X = H; 65% yield), 15% of 2-*o*-aminophenylbenzothiazole[5] (V, X = H). The formation of the latter can be explained through the nitrene (VI, X = H), in the triplet state, obtained by the loss of a molecule of nitrogen from III (X = H). In order to minimise the formation of by-products during the heterocyclisation, the azido compound was heated in toluene. Even after prolonged refluxing in this solvent no decomposition was observed. However, refluxing in xylene solution for four hours or heating a decalin solution at 150–55° for 45 minutes gave 75–80% yield of IV (X = H), exclusively. Thus, heating in neutral solvents around 150° appears to be the optimum condition for the heterocyclisation reaction. Adopting this procedure, three 2-*o*-azidophenylbenzothiazoles with a substituent in 6-position have been cyclised to the corresponding indazolo (2, 3-*b*)-benzothiazoles.

The fact that none of these 2-*o*-azidophenyl benzothiazoles decompose below 140° favours a stepwise mechanism and not a concerted one. As a means of obtaining 2-arylindazoles (VII), all the indazolo (2, 3-*b*) benzothiazoles (IV) have been subjected to desulphurisation by refluxing with Raney Ni in alcohol for 6 hours. In all these cases the expected 2-arylindazoles have been obtained in excellent yields and in pure state.

After the completion of this work, the publication of Suschitzky *et al.*[6], which reported the conversion of 2-*o*-azidophenylbenzothiazole to III (X = H) came to our notice. Neither the method of preparation of III (X = H) nor the physical constants of III and IV (X = H) were mentioned in this report. They obtained 2-phenyltetrahydroindazole on desulphurisation of IV (X = H) in xylene. This result, in contrast to the one obtained in the present work, appears understandable in view of the drastic conditions employed.

Application of this method, for the synthesis of other fused heterocycles from 2-*o*-azidoarylazoles is in progress.

ACKNOWLEDGEMENT

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1. Smith, P. A. S., In : *Nitrenes,* Edited by W. Lwowski, Interscience, 1970.
2. Stevens, H. N. E. and Stevens, M. F. G., *J. Chem. Soc.,* (*C*), 1970, p. 2308.
3. Bellamy, L. J., *The Infrared Spectra of Complex Molecules,* John Wiley, 1960, p. 267.
4. Abramovitch, R. A. and Adams, K. A. H., *Can. J. Chem*, 1961, 39, 2516.
5. Hein, D. W., Alheim, R. J. and Leavitt, J. J., *J. Am. Chem Soc.,* 1957, 79, 427.
6. McRobbie, I. M., Meth-Cohn, O. and Suschitzky, H., *Tetrahedron Letters,* 1976, p. 929.

COPPER (II)-ETHYLENEDIAMINE-SULFADRUGS COMPLEXES

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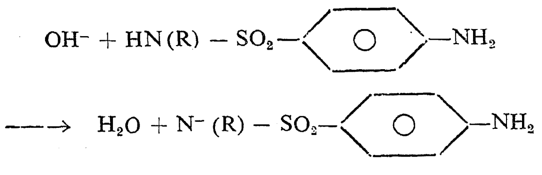
ABSTRACT

Cupper (II) complexes of the composition $Cu(en)_2D_2$ Where en = ethylenediamine and HD = sulfadrugs, *viz.*, sulfathiazole, sulfapyridine, sulfamerazine or sulfadiazine, have been prepared and characterized by analysis, infrared, electronic spectral and magnetic data. The complexes are planar and paramagnetic and the sulfadrug molecules act as anions.

INTRODUCTION

REACTIONS of the sulfadrugs with Zn (II), Cd (II), Hg (II)[1] and Cu (II)[2] have been successfully carried out using high concentrations of metal salts. In alkaline medium, however, the –NH– protons of the sulfadrug molecules are expected to be labile and the drugs exist in the anionic form.



In order to investigate such a behaviour of the sulfadrugs, the present study has been undertaken and some mixed Cu (II) compelxes of sulfadrugs and ethylenediamine are reported.

EXPERIMENTAL

The complexes were prepared by reacting solution of copper (II) chloride (10 m moles) or Copper (II) hydroxide (10 m moles) and ethylenediamine (2 ml) in the minimum amount of water (5 ml) with hot ethanolic solution (25 ml) of sulfadrug (20 m moles) and ethylenediamine (2 ml). The resulting solids were obtained on keeping the reactants for 1 hr., filtered and washed with 50% alcohol and dried at 110° C.

Metal part and sulphur in the complexes were estimated gravimetrically and nitrogen was analysed by micro-analytical technique using a coleman N-analyzer. Infrared and electronic spectra were recorded on Perkin–Elmer–257 and a Cary–14 model respectively. Magnetic measurements were made on Faraday Balance (Cahn, Magnetic susceptibility Apparatus) using $Hg\ [CO\ (NCS)_4]$ as calibrant. Diamagnetic currections were applied using

TABLE I

Analytical, magnetic and electronic spectral data of Copper (II)-ethylenediamine-sulfadrug complexes

Compounds	Melting pt. decompn. pt.	Analysis (%)* Metal	Nitrogen	Sulphur	Magnetic moment (μ_{eff}) B.M. temp. 19° C	Electrnoic spectral. λ_{max} mμ
1. Cu (*en*)$_2$ (ST)$_2$	184° C ST –200° C	8·98 (9·09)	20·30 (20·20)	18·23 (18·47)	1·82	210, 270, 300, 590 ST–250, 310
2. Cu (*en*)$_2$ (SP)$_2$	216° C Sp –191° C	9·10 (9·25)	20·22 (20·56)	9·18 (9·39)	2·05	215, 275, 305, 570 SP–240, 311
3. Cu (*en*)$_2$ (SM)$_2$	212° C SM–234° C	8·66 (8·85)	23·69 (23·63)	8·77 (9·00)	1·92	222, 260, 305, 555 SM–270, 305
4. Cu (*en*)$_2$ (SD)$_2$	213° C SD –253° C	9·26 (9·22)	24·76 (24·59)	9·04 (9·37)	1·99	220, 265, 310, 560 SD–292, 318, 332
5. Cu (*en*)$_2$ (ST)$_2$**	185° C ST –200° C	8·88 (9·09)	20·47 (20·20)	18·50 (18·47)	2·10	215, 270, 300, 585 ST–250, 310
6. Cu (*en*)$_2$ (SP)$_2$**	217° C SP –191° C	9·18 (9·25)	20·70 (20·56)	9·59 (9·39)	1·99	215, 272, 305, 570 SP–240, 311
7. Cu (*en*)$_2$ (SM)$_2$**	214° C SM–234° C	8·68 (8·85)	23·58 (23·63)	9·32 (9·00)	1·93	220, 255, 305, 550 SM–270, 305
8. Cu (*en*)$_2$ (SD)$_2$**	215° C SD –253° C	9·14 (9·22)	24·35 (24·59)	9·73 (9·37)	1·88	225, 265, 305, 555 SD–292, 318, 332

* Figures in parentheses are calculated values.

** Prepared from Cu (OH)$_2$, others from CuCl$_2$.

Abbreviations: HST = Sulfathiazole, HSP = Sulfapyridine; HSM = Sulfamerazine, HSD = Sulfadiazine.

Pascal's constants. The analytical data and some general characteristics of the complexes are summarized in Table I.

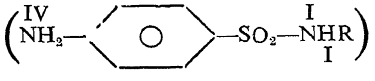
RESULTS AND DISCUSSION

All the Cu (II) complexes, presently reported, are either blue or bluish-violet in contrast to green Cu (II) complexes of the sulfadrugs reported earlier[2]. All the complexes are insoluble in water and common organic solvents and melt with decomposition in the range 180–220°. The course of reaction may be as follows :

$$HD + en \longrightarrow [enH^+]\, D^-$$

$$Cu + 2\, en + 2\, D^- \longrightarrow Cu\,(en)_2\, D_2$$

The effect of pH on the UV absorption bands of the sulfadrugs



appearing due to sulfanilamide group and the substituent at N has been studied by Vandenbelt and Doub[3]. The acidic ionization (in alkaline solution) results in a shift of sulfanilamido band (240–270 mμ) towards shorter wavelength with a little change in intensity while, basic dissociation (in acidic solution) decreases the intensity almost to disappearance of the band. In the present complexes, isolated from the basic medium of ethylenediamine, the shift of the characteristic sulfanilamide band to shorter wavelengths indicates the presence of drug anion as above. The absorption bands due to substituents at N^1, *viz.*, thiazole in sulfathiazole, pyridine in sulfapyridine, pyrimidine in sulfadiazine and 4-methyl pyrimidine in sulfamerazine, observed between 280–320 mμ, similarly shift to shorter wavelengths.

In octahedral Cu (II) complexes, a band around 800 mμ due to $^2T_{2g} \leftarrow {}^2E_g$ transition[4] blue-shifts considerably due to Jahn–Teller effect as the stereochemistry of Cu (II) complexes departs from octahedral to square planar. In general, the amine complexes of Cu (II) are blueish-violet and have been shown to have planar geometry. Cu $(en)_2^{2+}$ complexes show a *d-d* transition band around 550 mμ (18·2 kK)[5] in solid state and 545 mμ in solution[6]. Cu $(en)_2$ $(C\ 10_4)_2$ and Cu $(en)_2$–$(SCN)_2$ are isostructural and square planar. They show similar visible absorption bands around 520 mμ (19·2 kK) and 515 mμ (19·4 kK) respectively in solid state[7]. Since Cu $(en)_2D_2$ complexes absorb between 550–590 mμ and are either blue or bluish-violet, they are predominantly planar or at the most 'pseudo-octahedral with weakly coordinated drug anion.

All the Cu $(en)_2D_2$ complexes are normal paramagnetic and the μ_{eff} lies between 1·80–2·10 BM in agreement with the observed values for one unpaired electron in Cu (II) complexes[8].

Infrared spectra of the complexes show several bands in the 3500–3000 cm^{-1} due to ν NH from drug and *en* molecules. Although one of the series of the complexes has been prepared from copper (II) hydroxide but the close similarity of i.r. spectra of copper (II) chloride complexes indicates absence of OH group. When ν NH, δ NH_2, phenyl ring, SO_2 asym., SO_2 Sym. and τ NH_2 vibrational modes of the sulfanilamide part[9] (appearing in the regions 3500–3000, 1660–1620, 1600–1500, 1350–1300, 1150–1120 and 680–670 cm^{-1} respectively) in the sulfadrugs and Cu $(en)_2D_2$ complexes are compared, after eliminating the bands due to ethylenediamine, no significant changes are observed. It appears that the electron density has uniformly distributed itself over the entire drug molecule. In the case of sulfamerazine and sulfadiazine complexes, however, the SO_2 asymmetric band shows lowering of $\sim$ 10 cm^{-1}, which may be due to the slight decrease of electron density over-SO_2-group.

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1. Narang, K. K. and Gupta, J. K., *J. Inorg. Nucl. Chem.*, 1976, 38, 589.
2. — and —, *Ind. J. Chem.*, 1975, 13, 705.
3. Vandenbelt, J. M. and Doub, L., *J. Am. Chem. Soc.*, 1944, 66, 1633.
4. Dunn, T. M., *The Visible and Ultraviolet Spectra of Complex Compounds in Modern Co-ordination Chemistry*, Edited by J. Lewis and R. G. Wilkins, Interscience, New York, 1960.
5. Jorgensen, C. K., *Absorption Spectra and Chemical Bonding in Complexes*, Pergamon Press Ltd., 1962, p. 286.
6. Ojima, H. and Sone, K., *Bull. Chem. Soc. Jap.*, 1962, 35, 298.
7. Cristini, A. and Ponticelli, G., *J. Inorg. Nucl. Chem.*, 1975, 35, 2696.
8. Cotton, P. A. and Wilkins, G., *Advanced Inorganic Chemistry*, Interscience, New York, 1966, p. 902.
9. Nakanishi, K., *Infrared Absorption Spectroscopy*, Nankodo Company, Ltd, 1962, p. 216.

EFFECT OF SUBSTITUTION AND ITS LOCATION ON LIQUID CRYSTALLINE PROPERTIES OF CHOLESTERYLBENZOATE AND CHOLESTERYL CINNAMATE

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ABSTRACT

Cholesteryl 2- and 3-methoxybenzoates are non-mesomorphic, cholesteryl 2-nitrobenzoate is monotropic smectic and enantiotropic cholesteric. Cholestery 4-nitrocinnamate exhibits enantiotropic smectic and cholesteric mesophases, whereas cholesteryl 4-chlorocinnamate is only cholesteric. The mesomorphic properties are explained by comparing these compounds with other related compounds.

THE detailed study of many mesogenic homologous series has helped to evolve some general rules for the effect of chemical constitution in the nematogenic and smectogenic compounds[1]. The effect of chemical constitution on cholesteric mesophase has been also reported recently[2,3]. Dave and Vora[4] and Barral *et al.*[5], have reported *ortho, meta,* and *para* substituted benzoates of cholesterol and some substituted benzoates and cinnamates of cholesterol, respectively. From the study of substituted benzoates of cholesterol[4] it was observed that effect of same substituent at different locations is quite interesting. Generally the order of thermal stability is para > parent compound > meta > ortho. All the substituted benzoates of cholesterol reported exhibit mesomorphism. Some more substituted benzoates and cinnamates of cholesterol are reported here which are compared with other known compounds. Transition temperatures of these compounds are recdorded in Table I.

Reference to Table I shows that methoxy group in the 4-position of cholesteryl benzoate, enhances cholesteric thermal stability quite appreciably but

TABLE I

Name of the Compound	Transition temperatures (°C)		
	Smectic	Choles-teric	Isotropic
1. Cholesteryl 4-methoxy benzoate[6]	..	180·0	268·0
2. Cholesteryl 3-methoxy-benzoate	..	..	153·0
3. Cholesteryl 2-methoxy-benzoate	..	..	122·0
4. Cholesteryl 2-nitro-benzoate[a]	(98·5)*	147·5	155·0
5. Cholesteryl 4-nitro-benzoate[4,7]	..	191·5	260·0
6. Cholesteryl 4-chloro-benzoate[4,5]	..	170·0	257·0
7. Cholesteryl 4-nitro-cinnamate	174·5	185·0	280·0 (decomp.)
8. Chalesteryl 4-chloro-cinnamate[b]	..	150·0	274·0
9. Cholesteryl benzoate[4]	..	150·0	178·0
10. Cholesteryl cinnamate[8]	..	160·5	215·0

* Values in the parenthesis indicate monotropy.

[a] Sandquist and Gorton report 150·9, 156·9[7].

[b] Barrall *et al.* report 146·0, 262·0[5].

in the 2- and 3- positions, the same group destroys the mesomorphism. This is quite interesting. Substituted benzoates of cholesterol so far reported, irrespective of the location of the substituent exhibit mesomorphism. Arora *et al.*[9], and Barrall *et al.*[5], have suggested with the help of molecular models that in the aromatic esters, due to the steric factors, twist would occur which would be dependent on the size of the substitutent in the adjoining phenyl ring. It would be reasonable to assume that the methoxy group due to its size and position would create steric hindrance in the molecule. Due to the steric hindrance, the benzene ring may not be able to assume any position in the plane of cholesterol molecule which ultimately destorys the mesomorphism.

Cholesteryl 2-nitrobenzoate exhibits monotropic smectic and enantiotropic cholesteric mesophases. Sandquist and Gorton report only cholesteric mesophase for this compound[7]. Cholesteryl 4- and 3-nitrobenzoates exhibit only cholesteric mesophase. The solid-cholesteric transition temperatures of cholesteryl 3- and 2-nitrobenzoates do not differ much. The smectic mesophase is observed in the 2-substituted compound as it could be supercooled sufficiently without crystallization to exhibit smectic mesophase. Compared to methoxy substituent, nitro substituent behaves differently in the 2- and 3-positions. This behaviour of nitro group is not surprising. Nitro group is known to enhance the thermal stability and the enantiotropy due to very high polarity of the group. Similar behaviour is exhibited by nitro group in other mesomorphic compounds[1,10].

Cholesteryl 4-nitrocinnamate exhibits smectic and cholesteric mesophases whereas cholesteryl 4-nitrobenzoate is only cholesteric. This is quite an interesting observation as difference between the two compounds is only a trans- —CH=CH— group. It would be difficult to postulate the appearance of smectic mesophase in a single compound by different modifications. $-NO_2$ group is known to promote smectogenic tendencies of a compound and more so when it does not increase the breadth of a compound[1]. Cholestericisotropic transition temperature of cholesteryl 4-nitrocinnamate is higher than that of cholesteryl 4-nitrobenzoate. This is due to the increased length and polarizability of the molecules in the cinnamate ester due to the presence of trans —CH=CH— group.

Cholesteryl 4-chlorocinnamate exhibits cholesteric mesophase. Thermal stability of this compound is more than that of cholesteryl 4-chlorobenzoate. However the smectic tendency is not promoted eventhough chloro group is also known to promote smectic tendencies in other compounds[1].

Above discussion shows that it would require some more data to evolve general rules for the effect of chemical constitution on cholesteric mesophase.

The compounds were prepared by the method reported earlier[4,8].

The author expresses his thanks to Professor S. M. Sethna, Dr. J. S. Dave and Professor K. N. Trivedi, for their interest in the work.

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1. Gray, G. W., *Molecular Structure and the Properties of Liquid Crystals,* Academic Press, New York, 1962.
2. Elswer, W., *Mol. Cryst.*, 1967, 2, 1.
3. Dave, J. S. and Vora, R. A., *Mol. Cryst. Liquid Cryst.*, 1971, 14, 319.
4. — and —, *Indian J. Chem.*, 1973, 11, 19.
5. Barrall, E. M., II, Bredfeldt, K. E. and Vogel, M. J., *Mol. Cryst. Liq. Cryst.*, 1972, 18, 195.
6. Dave, J. S. and Vora, R. A., *Liquid Crystals and Ordered Fluids*, Johnson, J. F. and Porter, R. S., Ed., Plenum Press, New York, 1970, p. 477.
7. Sandquist, H. and Gorton, J., *Ber. dt. Chem. Ges.*, 1930, 63, 1759; *Chem. Abstr.*, 1930, 24, 5042.
8. Dave, J. S. and Vora, R. A., *Mol. Cryst. Liq. Cryst.*, 1971, 14, 319.
9. Arora, S. L., Fergason, J. L. and Taylor, T. R., *J. Org. Chem.*, 1970, 35, 4055.
10. Dave, J. S. and Dewar, M. J. S., *J. Chem. Soc.*, 1954, p. 4616; 1955, p. 4305.

STUDIES ON THE COMPLEXES OF PENTACYANOAMMINEFERRATE (II) AND SOME ORGANIC BASES

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ABSTRACT

Sodium pentacyanoammineferrate (II) reacts with *p*-phenylenediamine, *p*-anisidine, *m*-toluidine and N-methylaniline to give greenish blue complexes. The composition is 2:1 with *p*-phenylenediamine and 1:1 with the other three bases, confirmed by chemical analysis. Infrared spectra of the complexes show that the base is coordinated to iron through the amino group. Magnetic measurements show that iron is in the + 2 oxidation state.

INTRODUCTION

THE color reaction between pentacyanoammineferrate (II) and (III) and some organic compounds like aromatic amines, aldehydes, thioaldehydes and hydrazines has been used for spot test analysis of these organic reagents[1-4]. While Herington[3] has carried out detailed studies on the reaction of pentacyanoammineferrate (III) with aromatic amines, very little has been done towards this end for pentacyanoammineferrate (II). Studies have thus been carried out on the reaction of pentacyanoammineferrate (II) with *p*-phenylenediamine, *p*-anisidine, *m*-toluidine and N-methylaniline.

EXPERIMENTAL

N-methylaniline, *p*-phenylenediamine, *p*-anisidine and *m*-toluidine were BDH products and were purified before use by distillation or crystallisation from ethanol. Sodium pentacyanoammineferrate (II) was prepared from sodium nitroprusside (AnalaR, BDH) by the method described by Brauer[5].

Bausch and Lomb Spectronic 20 Spectrophotometer and Elico (India) pH meter, LI–10, were used.

Infrared spectra were recorded in KBr discs using Beckman IR '20' spectrophotometer. Magnetic measurements were carried out by Guoy's method at room temperature (30 ± 1° C).

Preparation of the Complexes :

N-methylaniline, *p*-phenylenediamine, *p*-anisidine and *m*-toluidine were converted into their respective hydrochlorides. To 50 ml of a solution of 0·01 *M* base hydrochloride (0·005 *M* in the case of *p*-phenylenediamine) was added 50 ml of 0·01 *M* pentacyanoammineferrate (II). After allowing to stand for 1 hour, a solution of zinc acetate (about 4 g) in dilute acetic acid (20 ml of glacial acetic acid in 100 ml of water) was added to precipitate the green gelatinous product. This was filtered off immediately and washed with water till free from acid and Zn^{++}. The product was further washed with alcohol, then ether and finally dried in vacuum over anhydrous calcium chloride.

The complexes were chemically analysed[6] for iron and zinc after decomposing by boiling with *aqua regia*. Carbon, hydrogen and nitrogen were estimated at I.I.P., Dehra Dun.

RESULTS AND DISCUSSION

The organic bases give bluish green complexes with pentacyanoammineferrate (II) in about 1 hour and the reaction decreased in the following order:

p-phenylenediamine > *p*-anisidine > *m*-toluidine > N-methylaniline.

Vosburg and Cooper's method showed the formation of only one complex species. The λ_{max} of these complexes were observed at 715, 750, 730 and 740 nm for *p*-phenylenediamine, *p*-anisidine, *m*-toluidine and N-methylaniline respectively. Job's method and molar ratio method showed the stoichiometric ratio of *p*-phenylenediamine and $Na_3\,[Fe\,(CN)_5\,(NH_3)]$ is 1 : 2 and of *p*-anisidine, *m*-toluidine and N-methylaniline is 1 : 1, which is confirmed by chemical analysis (Table I).

The pH of 0·01 *M* pentacyanoammineferrate (II) was 9·7 and that of *p*-phenylenediamine hydrochloride, *p*-anisidine hydrochloride, *m*-toluidine hydrochloride and N-methylaniline hydrochloride of the same concentration was 2·9, 4·5, 4·0 and 4·3 respectively. After mixing, the pH rises almost immediately (within 30 seconds) to 6·5 in all the cases and becomes constant within one hour at a pH of 8·0, 7·8, 7·3 and 7·2 respectively. The rise in pH is obviously due to the release of ammonia according to the following scheme :

$$\begin{aligned}&\text{Base} + Na_3\,[Fe\,(CN_5)\,(NH_3)] \longrightarrow \\ &\quad Na_3\,[Fe\,(CN)_5\,(\text{Base})] + NH_3\end{aligned}$$

Magnetic measurements reveal their diamagnetic character, showing that iron is present as Fe^{2+} in all the complexes. The infrared bands together with their assignments are given in Table II. The band appearing at 3300 cm^{-1} is due to the amine group of the base[7] (absence of a band around 1600 cm^{-1} rules out water of crystallisation or NH_3). The C–N band appearing around 1250 cm^{-1} also shows shifts indicating coordination through the amine group[8]. The C $\equiv$ N stretching frequency

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TABLE I
Chemical analysis of the complexes

Complex		% Zn	% Fe	% C	% N	% H
$Zn_3 [Fe(CN)_5NH_2C_6H_4NH_2Fe(CN)_5]$	Found	27·52	16·30	29·10	23·39	1·00
	Calc.	28·93	16·51	28·31	24·77	1·18
$Zn_3 [Fe(CN)_5NH_2C_6H_4OCH_3]_2$	Found	24·80	13·53	36·13	19·94	1·00
	Calc.	24·06	13·76	35·38	20·64	1·10
$Zn_3 [Fe(CN)_5NH_2C_6H_4CH_3]_2$	Found	26·03	14·43	37·59	21·99	1·00
	Calc.	25·05	14·32	36·83	21·48	1·15
$Zn_3 [Fe(CN)_5NHCH_3C_6H_5]_2$	Found	25·75	14·41	35·97	22·32	1·00
	Calc.	25·05	14·32	36·83	21·48	1·15

TABLE II
Infrared absortion bands (cm^{-1})

Complex	$\nu(NH_3)$* or stretching frequency due to amine	$\nu(C\equiv N)$	$\nu(C—N)$	$\nu(Fe—C)$	$\nu(Fe—N)$
$Na_3 [Fe(CN)_5(NH_3)] \cdot 3H_2O$	3400* b, st	2040 s, st	..	570 s, st	490 s, m
Complex of pentacyanoammine feerrate (II) with					
p-phenylenediamine	3300 b, st	2060 s, st	1320 sh	565 s, st	470 b, st
p-anisidine	3300 b, st	2090 s, st	1250 s, st	570 m	500 w
m-toluidine	3300 b, st	2080 s, st	1255 m	580 m	500 w
N-methylaniline	3300 b, st	2070 s, st	1260 m	575 m	500 w

s–sharp; st–strong; b–broad; m–medium; w–weak.

appears around 2080 cm^{-1}. The band around 600 cm^{-1} and 500 cm^{-1} may be assigned to Fe–C and Fe–N respectively. It is thus evident that iron in these complexes is coordinated to cyano groups and to the organic base through nitrogen of the amine group.

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1. Anger, *Mikrochim. Acta (Wein)*, 1937, 2, 3.
2. Feigl, F., *Spot Tests in Organic Analysis*, Elsevier, New York, 1956.
3. Herington, E. F. G., *J. Chem. Soc.*, 1956, p. 2747 ; 1958, p. 4683 ; 1958, p. 4771 ; 1959, p. 3633.
4. Jaselkis, B., *J. Amer. Chem. Soc.*, 1961, 83, 1082.
5. Brauer, G., *Hand-Book of Preparative Inorganic Chemistry*, Academic Press, New York, 1965, 2, 1509.
6. Vogel, A. I., *A Text-Book of Quantitative Inorganic Analysis*, Longmans Green, London, 1961.
7. Rao, C. N. R., *Chemical Applications of Infrared Spectroscopy*, Academic Press, New York, 1963.
8. Nakamoto, K., *Infrared Spectra of Inorganic and Coordination Compounds*, John Wiley, New York, 1962.

PHYSIOLOGICAL STUDIES ON THE EFFECTS OF NUTRITIONAL IMBALANCE ON THE CENTRAL NERVOUS SYSTEM: EFFECTS OF THIAMINE-DEFICIENCY ON SOME KINETIC CHARACTERISTICS OF ACETYLCHOLINESTERASE IN THE DIFFERENT REGIONS OF THE BRAIN OF CHICKEN, *GALLUS DOMESTICUS*

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Abstract

The Michaelis constants for acetylcholinesterase (E.C. 3·1·1·7, AchE) were determined in the cerebrum, cerebellum, optic lobes and medulla oblongata of normal and thiamine-deficient chicken, *Gallus domesticus*. In general the cerebellar AchE has lower Km and lower Vm on inducing thiamine deficiency. AchE in the optic lobes had higher Km and higher Vm (over controls) in the thiamine deficient birds. However, brain stem exhibited lower Km and lower Vm during thiamine deficiency. The significance of the changes observed is discussed.

CONSIDERABLE information is available on enzymatic activities in vertebrates during thiamine deficiency[1-6]. It has been demonstrated that the absence of thiamine affects the distribution of thiamine dependent enzymes that influence carbohydrate metabolism in birds and mammals[1,3]. Since the oxidative catabolism of lactate and pyruvate is essential for the normal activity of brain tissue, and since pyruvate is an important source of acetyl group of acetylcholine[4], an interference with the oxidative decarboxylation of pyruvic acid during thiamine deficiency[5] may be expected to produce abnormalities of neuronal activity[4]. However, information on the central nervous system (CNS) activity and function during thiamine deficiency is lacking. Motivated by this consideration and keeping in view the fact that AchE activity is related to the electrical activity[7], and electrical activity to the potentiality of cell response[8], the present study was undertaken.

Material and Methods

Two days old white leg-horn chicken, *Gallus domesticus* (10–12 g) were reared in the laboratory at 36 ± 2° C. The controls were fed on standard chicken feed. The experimental birds were fed on polished rice to induce thiamine deficiency[1]. Water was available *ad libitum.*

The normal and thiamine deficient chicken were decapitated after 25 days. The brain was dissected and kept with Ringer[10] at 0° C. The different regions of the brain (cerebrum, cerebellum, optic lobes and medulla oblongata) were separated with sterilized instruments, weighed in ice-cold Ringer and immediately used for analysis.

The activity level of AchE was determined spectrophotometrically by the method of Hestrin[9]. The mean values of AchE from 5 different tissues (obtained from 5 chickens) of cerebrum, cerebellum optic lobes and medulla oblongata were plotted as (1/v) against (1/S) where [(v)] is the reaction velocity and [(S)] the substrate concentration as per the method of Lineweaver and Burk[11]. The slopes, Vm, and Michaelis constants (Km) were calculated by the method of least squares.

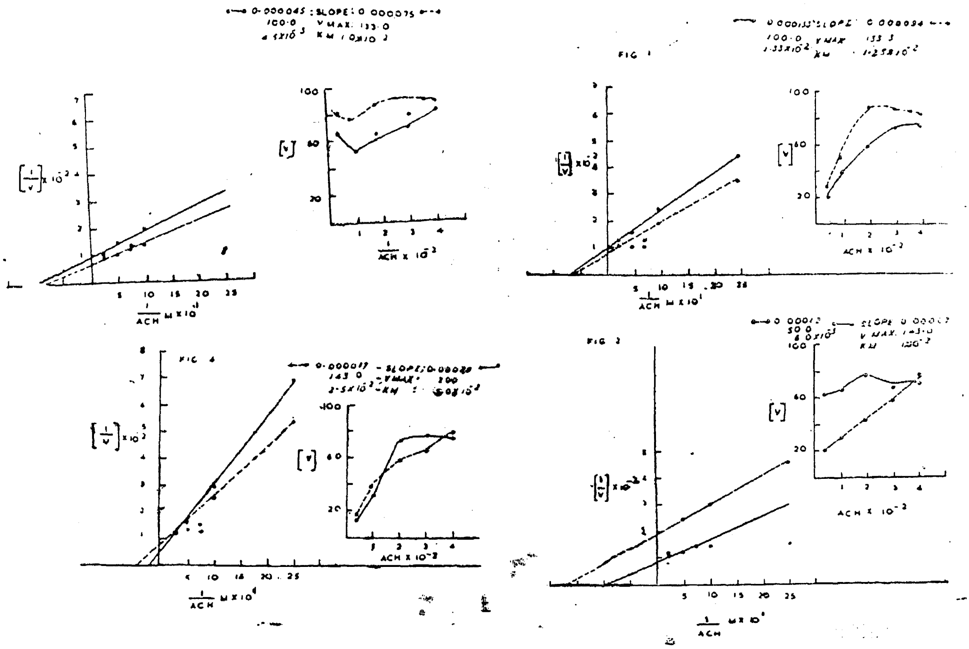
Results and Discussion

The results are summarized in Figs. 1–4.

The activity levels of AchE in general showed an increase in the compartments of the brain studied, *viz.*, cerebrum, cerebellum, optic lobes and medulla oblongata during thiamine deficiency. It is also seen (Figs. 1–4) that the increase in the levels of AchE activity at different substrate concentrations showed some variation in relation to the region of the brain. Such an increase in different regions in the brain of the hypovitaminous chicken may either be due to the activation of enzyme synthetic machinery by the altered cellular environment prevailing in the brain or due to an increase in the rate of enzyme synthesis.

In the present study the Km values of AchE in the different regions of the brain of normal and thiamine deficient chicken were found varying. This suggests that the enzyme substrate affinity is not identical for all the regions and for the same region in thiamine deficiency. One should expect decreased enzyme substrate affinity as a consequence of high ES dissociation velocity or a relatively low Km values in the case of AchE of optic lobes and of cerebellum in thiamine deficient birds. The slope of the curve (Km/V) bears a proportionate correlation with the present alteration of enzyme reaction velocities. The AchE of medulla oblongata had higher V_{max} in control and also in thiamine deficient animals than the activity levels of AchE in other regions of brain (Figs. 1–4).

The comparison of v and S reveals a steeper curve for cerebral AchE of normal and thiamine deficient chicken than the AchE of other regions, thus manifesting the sensitive affinity (Fig. 1). The kinetic characteristics of AchE show greater efficiency of cerebellar AchE than the AchE of other regions investigated, with Km value less than the other regions in thiamine deficiency. But in the controls, the kinetic characteristics of AchE show greater efficiency of AchE of optic lobes than the AchE of other regions with Km value less than the other regions. Thus, the substrate kinetic characteristics of AchE of control and experiment



FIGS. 1–4. Line weaver and Burk plots along with typical (V) *versus* (S) plots for acetylcholinesterase of cerebrum (Fig. 1), cerebellum (Fig. 2), optic lobes (Fig. 3) and medulla oblongata (Fig. 4). Each point in the plots is a mean of 5 observations made utilizing tissues from 5 chicken. The assay mixture contained 0·5 ml of 2% homogenate, 0·5 ml of buffered substrate (0·1 N NaCl + 0·02 M $MgCl_2$ + Acetylcholine chloride at different molar concentrations + 0·005% gelatin in 0·02 M phosphate buffer of pH 7). The mixture was incubated at 25° C for 2 minutes and the reaction was stopped by adding 1·0 ml of alkaline hydroxylamine hydrochloride solution (1 : 1 mixture of 2·0 M hydroxylamine hydrochloride and 3·5 N sodium hydroxide). After an additional one minute, 0·5 ml of 4 N hydrochloric acid was added followed by 0·5 ml of 0·37 M ferric chloride (in 0·1 N hydrochloric acid) solution. The optical density of the final solution was read in a DU-2 Beckman Spectrophotometer at 540 nm against distilled water. A blank was prepared in the same manner excluding the enzyme. Enzyme activity is expressed as micromoles of Ach hydrolyzed/mg/minute. O - - - - - - O Thiamine deficient ; O ———— O Controls.

groups demonstrate that during thiamine deficiency AchE becomes less efficient as indicated by the elevated Km values (Figs. 1–4) in optic lobes during thiamine deficiency and hence the active sites are affected in this compartment of the brain during thiamine deficiency. Whereas in the cerebrum, cerebellum and brain stem, the decreased Km indicates that the enzyme is more efficient and the active sites are not affected during thiamine deficiency in these compartments of the brain.

However, in general the kinetic characteristics of AchE of experimental group show greater emphasis towards the first order reaction pattern where maximal velocity is obtained, while the AchE of control group represents a tendency towards the zero order reaction pattern on the lines suggested by Neurath and Schwert[12] at relatively low S levels.

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1. Peters, R. A., *Biochemical Lesions and Lethal Synthesis* (Pergamon Student Editions), International Series of Monographs on Pure and Applied Biology : Div. Modern Trends in Physiological Sciences, 1963, p. 8.
2. Takahashi, Kisabaro, Nakamura, A. and Yoshitsugu, *J. Vitaminol.* (Tokyo), 1971, **17** (4), 207.
3. Pincus, J. H. and Wells, K., *Exp. Neurol.*, 1972, **37** (3), 495.
4. Fruton, J. S. and Simmonds, S., *General Biochemistry*, Asian Student Ed., 1965.
5. Nalini, U. P. and Nayeemunnisa, *Experientia*, 1976, **32** (2), 198.
6. Bamji, S. M. *Ind. J. Vitamin. Nutr. Res.*, 1972, **42**, 184.
7. Muralikrishnadas, P. and Venkatachari, S. A. T., *Life Sciences*, 1968, **7**, 617.
8. Sanvar Basha, S. and Swami, K. S., *Ann. Allergy*, 1968, **26**, 601.
9. Hestrin, S., *J. Biol. Chem.*, 1949, **180**, 249.
10. Prosser, C. L. and Brown F. A., *Comparative Animal Physiology*, Saunders Company, Philadelphia, 1961.
11. Lineweaver H. and Burk, D., *J. Am. Chem. Soc.*, 1934, **56**, 658.
12. Neurath, H. and Schwert, G. W., *Chem. Rev.*, 1950, **46**, 69.

LETTERS TO THE EDITOR

PATHOLOGIES IN INTERACTING MASSIVE VECTOR AND SCALAR FIELDS

FIELD theory of high spin particle interactions suffers from troubles of various sorts both at the quantized and classical levels. Johnson-Sudarshan effect[1], causality violation[2,3], Lorentz non-covariance[4] and breakdown of constraints[5] are some of the pathologies afflicting high spin field theories. We here report the results of a study concerning the possibility of causality violation and breakdown of constraints in the mutual interaction between a spin one field and a spin zero field under several model coupling schemes. Apart from its pedagogic value this investigation brings to light certain curious but mild forms of pathology which do not seem to have been noted before.

We first consider the simplest coupling of a massive neutral vector field to a complex scalar field with an interaction Lagrangian of the form

$$a_I = ig\,[(\partial_\mu \phi^\dagger)\phi - \phi^\dagger \partial_\mu \phi]\,A_\mu . \qquad (1$$

The equations of motion that follow from this are :

$$(\partial^2 - m^2)A_\nu - \partial_\nu \partial_\lambda A_\lambda = -ig\,[(\partial_\nu \phi^\dagger)\phi - \phi^\dagger \partial_\nu \phi] \qquad (2)$$

$$(\partial^2 - \mu^2)\phi = ig\,[2(\partial_\lambda \phi)A_\lambda + \phi\partial_\lambda A_\lambda] \qquad (3)$$

$$(\partial^2 - \mu^2)\phi^\dagger = -ig\,[2(\partial_\lambda \phi^\dagger)A_\lambda + \phi^\dagger \partial_\lambda A_\lambda] \qquad (4)$$

Equation (2) is not a true equation of motion since it does not contain the second time derivative of A_4. The constraint in (2) is eliminated by taking the divergence of eq. (2) and making use of eqs. (3) and (4). This gives the secondary constraint :

$$\partial_\nu A_\nu = \frac{2g^2}{m^2}[\phi^\dagger \phi \partial_\nu A_\nu + (\partial_\nu \phi^\dagger)\phi A_\nu + \phi^\dagger(\partial_\nu \phi)A_\nu]. \qquad (5)$$

When $2g^2/m^2\,\phi^\dagger\phi \neq 1$ we can substitute eq. (5) in eq. (2) to get the constraint-free equation

$$(\partial^2 - m^2)A_\nu - \frac{2g^2}{m^2}[\phi^\dagger\phi\,\partial_\nu\partial_\lambda A_\lambda + \partial_\mu A_\mu \partial_\nu(\phi^\dagger\phi) + \partial_\nu\{(\partial_\mu \phi^\dagger)\phi A_\mu + \phi^\dagger \partial_\mu \phi A_\mu\}] + ig\,[(\partial_\nu \phi^\dagger)\phi - \phi^\dagger \partial_\nu \phi] = 0 \qquad (6)$$

When the scalar field assumes a value for which $\phi^\dagger\phi = m^2/2g^2$ the secondary constraint relation breaks down. However, it can easily be shown that the loss of constraint is a Lorentz-covariant phenomenon and consequently our theory does not exhibit the type of non-covariance observed by Jenkins[5].

The characteristic determinant D (n) of the system of eqs. (6), (3) and (4) is found by replacing $\partial_\mu \to n_\mu$ in the second derivative terms and evaluating the determinant of the coefficients of A_μ, ϕ and $\phi^\dagger$.

$$D(n) = (n^2)^6\left(1 - \frac{2g^2}{m^2}\phi^\dagger\phi\right) \qquad (7)$$

Normals to the characteristic surfaces are determined by setting D (n) = 0. We see that the propagation in causal unless $\phi^\dagger\phi = m^2/2g^2$ and at this particular value of the scalar field the nature of propagation is undetermined in that normals can be space-like, light-like or time-like. We note that it is precisely at this value that the secondary constraint breaks down.

Setting up the quantized version of the above theory in the usual fashion we observed that in the S-matrix in interaction picture normal-dependent terms survive and hence the quantized theory is non-covariant. This result may be taken as an elaboration of the Jenkins conjecture[4] that a theory manifesting causality violation at the classical level is non-covariant at the quantized level.

Modifying the interaction Lagrangian (1) to the form

$$a_I = ig\,[(\partial_\mu \phi^\dagger)\phi - \phi^\dagger \partial_\mu \phi] - g^2 \phi^\dagger \phi A_\mu A_\mu \qquad (8)$$

we find that in the resulting theory field propagation is causal and there is no constraint breakdown at any value of the scalar field. The corresponding quantized theory is Lorentz-covariant also.

Proceeding, in exactly the same way as above, we can establish analogous results for the interactions between neutral vector and neutral scalar field, complex vector and neutral scalar field and complex vector and complex scalar fields.

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1. Johnson, K. and Sudarshan, E. C. G., *Ann. Phys.*, 1961, 13, 126.
2. Velo. G. and Zwanziger, D., *Phys. Rev.*, 1969, 186, 1337.
3. — and —, *Ibid.*, 1969, 188, 2218.
4. Jenkins, J. D., *J. Phys. A*, 1973, 6, 1935.
5. —, *Ibid.*, 1974, 7, 1129.

LOW TEMPERATURE MAGNETIC SUSCEPTIBILITY OF THULIUM SESQUIOXIDE

STUDY of rare-earth sesquioxide has been a subject of considerable interest in recent years. All rare-earths form sesquioxides where their ions are presumed to exist in trivalent state. Antiferromagnetic ordering have been observed[1] [2] in some of these oxides (Tb_2O_3 Er_2O_3, Yb_2O_3 and Dy_2O_3) at low temperatures. To the best of our knowledge the magnetic studies of holmium and thulium and sesquioxide have not yet been reported in literature. In this note we report the magnetic susceptibility and magnetization of Tm_2O_3 from 4·2° K to 270° K. No magnetic ordering have been observed in this compound upto 4·2° K.

Tm_2O_3 with a stated purity of 99·9% was procured from Fluka AG Switzerland. It has cubic (C-type) structure, with room temperature cell parameter a = 10·488 Å. It belongs to space group T_h^7 (Ia3) and has 16 molecules per unit cell[3].

The magnetisation of the sample was measured using Foner magnetometer. In this technique sample vibrates perpendicular to a constant and homogeneous magnetic field direction, and the alternating signal induced in a pair of pick up coils is measured and compared to that from a nickel sample. The magnetization of the sample (1 to 50 mg) could continuously be recorded from 4·2 to 300° K at the magnetic field from 0 to 10 kgauss.

The results are shown in Fig. 1. The χ^{-1} *vs* T curve is a good straight line up to the lowest temperature and thus obeys the Curie Weiss law with Curie constant **C = 13·51° K/g. mole and paramagnetic Curie temperature $\theta = -20°$ K.** The only magnetic ion in this sample is Tm^{3+}. In the ground state[4] it has J = 6, L = 5, S = 1. This gives a magneton number $[p^2 = g\,J\,(J\ 1)]$ for this ion as 7·57. Experimental value of p can be obtained from the value of C using the relation[4]

$$p = (3kC/2N\beta^2)^{1/2} \qquad (1)$$

where k is Boltzmann constant, N the Avogadro number and β the Bohr magneton. The number 2 is coming in denominator because each molecule contains two Tm^{3+} ions. On computation one gets the experimental value of $p = 7{\cdot}35$. This value agrees fairly well with the theoretical value.

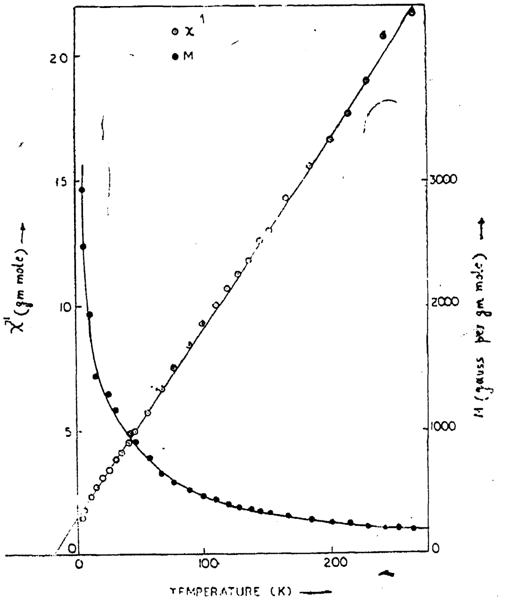

FIG. 1. The magnetisation (M) and the inverse magnetic susceptibility (χ^{-1}) *vs* temperature (°K) of the powdered sample of Tm_2O_3.

All the rare-earth sesquioxides studied are antiferromagnetic and hence it is expected that Tm_2O_3 may also be antiferromagnetic. The Curie-Weiss behaviour of χ supports this inference. However a negative value of θ and Curie-Weiss law behaviour can also come purely due to crystal field effect, whose contribution is larger than KT below 100° K even for the cubic symmetry[4]. Thus Curie-Weiss behaviour observed may be the combined effect of crystal field and antiferromagnetic exchange interaction. It can be said with certainty that Tm_2O_3 does not exhibit any magnetic order upto 4·2° K.

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1. Moon, R. M., Child, H. R., Kcchler, W. C. and Raubenheimer, J. L., *J. Appl. Phys.*, 1967, **38**, 1383.
2. —. — and —, *Phys. Rev.*, 1968, **176**, 722.
3. Wyckoff, R. W. G., *Crystal Structure*, Vol. II, John Wiley and Sons, New York, 1964, p. 5.
4. Martin, D. H., *Magnetism in Solids*, Illiffe Books Ltd., London, 1967.

INFRARED SPECTRAL STUDIES OF N-PHENYL AND N, N′-DIPHENYLTHIOUREA COMPLEXES WITH d^{10} METAL IONS

Introduction

THIOUREA class of compounds has not been studied extensively except for the first member, thiourea[1,2]. The complexes of thiourea (tu) with Zn (II), Cd (II) and Hg (II) chlorides have already been reported and their infrared spectra studied by Swaminathan and Irving[1]. There are reports about the synthesis and infrared spectral studies related to complexes of thioureas with Ni (II)[2], Co (II)[3], Fe (III)[4], Mn (II)[5] and Pd (II)[6]

The monosubstituted N-phenyl thiourea (Ptu) and the symmetrically disubstituted N, N-diphenylthiourea (DPtu) complexes with Zn (II), Cd (II) and Hg (II) chlorides have been investigated by us. Amongst these complexes, Ptu and DPtu complexes with $ZnCl_2$ and $HgCl_2$ are reported recently[7]. The composition of the complexes is different and the structural assignments are based on the assignments of Yamaguchi[6].

Experimental

Metal chlorides and other chemicals were of Analar grade. The complexes were prepared by treating the hot metal chloride solution in methanol with a slight excess of hot Ptu solution in the same solvent. The solution was concentrated and cooled in ice. The product was washed with methanol and dried in vacuum.

The above procedure was followed for DPtu complexes, but the complexes crystallise out on concentrating the solution. The complexes were analysed for metal, chloride, nitrogen and sulfur by standard procedures. The analytical data are given in Table I.

The infrared spectra of the samples were recorded in nujol mull on a Carl-Zeiss UR 10 Spectrophotometer from 4000–400 cm^{-1} and Beckman I.R. 12 Spectrophotometer from 400–200 cm^{-1}.

Results and Discussion

The complexes have 1 : 2 stoichiometry and they are colourless and amorphous. Jensen and Nielsen[8] by comparing the infrared spectra of thiourea with selenourea have assigned a band 630 cm^{-1} to C–S stretching. In Ptu and DPtu a band at 638 cm^{-1} and 630 cm^{-1} respectively could be assigned to CS stretching (Table I). The ν CS (stretching frequency) is reduced by 20–30 cm^{-1} in the DPtu complexes as in Ptu complexes of Zn (II), Cd (II) and Hg (II). This observation reveals the coordination of substituted thiourea with metal ions through sulphur.

The far infrared spectra of the above metal complexes contain medium strong to strong bands in the region 210 to 290^{-1} which indicate the existence of M–S band (Table I). These are clearly distinguished from M–Cl stretching vibrations found in the region 270 to 340 cm^{-1}. The band splitting for ν (Zn–Cl) indicates the increased covalency of the bond compared to that of other metal complexes. The appearance of only one band in the case of ($HgDPtu_2Cl_2$) and ($ZnPtu_2Cl_2$) indicates vibrational coupling of M–S and M–Cl stretching frequencies. Since divalent Zn, Cd and Hg have strong tendency to form tetrahedral complexes, the complexes presently discussed may have tetrahedral stereochemistry.

TABLE I

Complex	Elemental Analysis				Infrared Frequencies		
	Metal %	Cl %	S %	N %	ν CS	ν M–Cl	ν M–S
Ptu	..	..	..	..	638 ms	..	..
$ZnCl_2$. 2Ptu	14·88 (14·84)	15·98 (16·08)	14·45 (14·55)	12·62 (12·72)	622 m	..	277 s 287 s
$CdCl_2$. 2Ptu	23·18 (23·06)	14·45 (14·55)	13·35 (13·13)	11·60 (11·49)	596 m	319 s	283s
$HgCl_2$. 2Ptu	34·67 (34·82)	12·39 (12·31)	11·00 (11·14)	9·65 (9·73)	592 s	327–331 mb	277 s
DPtu	..	..	..	..	630 s	..	..
$ZnCl_2$. 2DPtu	10·90 (11·03)	12·13 (11·96)	10·77 (10·81)	9·48 (9·45)	610 ms	315 ms 328 ms	290 s
$CdCl_2$. 2DPtu	17·40 (17·58)	11·19 (11·09)	9·81 (10·01)	8·75 (8·76)	601 s	273 s	238 s
$HgCl_2$. 2DPtu	27·60 (27·54)	9·80 (9·74)	8·88 (8·81)	7·66 (7·69)	600 mw	..	261 s

The values in the parenthesis are the calculated values.
s = Strong, ms = Medium Strong, w = Weak, mb = Medium Broad, mw = Medium Weak.

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1. Swaminathan, K. and Irving, H. M. N. H., *J. Inorg. Nucl. Chem.*, 1964, **26**, 1291.
2. Gosavi, R. K. and Rao, C. N. R., *Ibid.*, 1967, **29**, 1937.
3. Moharana, S. N. and Dash, K. C., *Ind. Jour. Chem.*, 1970, **8** (II), 1023.
4. Birchall, T. and Morris, M. F., *Can. Jour. Chem.*, 1972, **80** (2), 201.
5. Stancheva Preya, *Nauch, Tv. Vissh. Pedagog. Inst. Plovdiv at Fiz., Khim Biol.*, 1970, **8** (1), 103; *C.A.* 1971, **74**, 60364 J.
6. Yamaguchi, A., Penland, R. B., Mizushima, S., Lane, T. J., Curran, C. and Quagliano, J. V., *J. Amer. Chem. Soc.*, 1958, **80**, 527.
7. Rath, P. C. and Mahapatra, B. K., *J. Ind. Chem. Soc.*, 1974, **51**, 710.
8. Jensen, K. A. and Nielsen, P. H., *Acta Chem. Scand.*, 1966, **20**, 597. and other references therein.

A CONVENIENT METHOD FOR THE PREPARATION OF α-ARYLAMINO PHENYL ACETIC ACIDS

McKenzie and coworkers[1] prepared α-phenylamino phenyl acetic acid by condensing α-helophenyl acetic acid with aniline. We have prepared α-arylamino phenyl acetic acid of the type C_6H_5·CH (R) COOH where R = phenylamino; *o*-, *m*- and *p*-tolylamino *o*- and *p*-anisylamino; *m*- and *p*-chlorophenyl amino; *p*-bromo and *p*-iodophenyl amino, etc. They have been prepared by hydrolysing α-arylamino phenyl acetonitriles, first by treating with concentrated H_2SO_4 as a result of which amide formation takes place which then gets hydrolysed by aqueous sodium hydroxide solution (10%) to the corresponding acids. This method is found to be convenient and the yield of the products is also very good.

Expertmental

(*a*) *Preparation of α-arylamino phenyl acetonitriles*

They have been prepared by the method suggested by Sandhu and coworkers[2]. The different substituents R, % yields and the m.p. in ° C are as follows:

Aryl, 85, 90°; *o*-tolyl, 79, 75°; *m*-tolyl, 82, 95°; *p*-tolyl, 80, 110°; *o*-anisyl, 85, 68°; *p*-anisyl, 86, 80°; *m*-chloro phenyl, 80, 78°; *p*-chloro phenyl, 78, 70°; *p*-bromo phenyl, 76, 100°; *p*-iodophenyl, 60, 75°. All compounds gave correct N-analysis.

(*b*) *Preparalion of α-arylamino phenyl acetamide*

α-Phenylamino phenyl acetonitrile (0·025 mole) was dissolved in concentrated sulphuric acid (10 ml) below 10° and kept for 48 hours at room temperature. Contents were then poured into ice-water and filtered. The filtrate was made alkaline by aqueous sodium hydroxide solution. The product obtained was crystallised from alcohol.

Different substituents R, % yield and the m.p. in ° C are as follows:

Aryl, 75, 120°; *o*-tolyl 63, 110°; *m*-tolyl, 60, 100°; *p*-tolyl, 65, 129°; *o*-anisyl, 72, 165°; *p*-anisyl, 74, 105°; *m*-chloro phenyl, 62, 103°; *p*-chloro phenyl, 58, 146°; *p*-bromo phenyl, 70, 174°; *p*-iodophenyl, 74, 172°.

All compounds gave correct N-analysis.

(*c*) *Preparation of arylamino phenyl acetic acid*

α-Arylamino phenyl acetamide (0·025 mole) was mixed with aqueous sodium hydroxide solution (10%) and refluxed for 4 hours. It was diluted with ice-water and filtered, The filtrate was then acidified with concentrated hydrochloric acid. The product obtained was crystallised from alcohol.

Different substituents R, % yield and the m.p. in °C are as follows:

Aryl, 55, 179° (Reported 175°); *o*-tolyl, 50, 149°; *m*-tolyl, 53, 145°; *p*-tolyl, 51, 164°; *o*-anisyl 49, 280° (*d*); *p*-anisyl, 51, 173°; *m*-chloro phenyl, 48, 147°; *p*-chloro phenyl, 45, 175°; *p*-bromo phenyl, 46, 171°; *p*-iodophenyl, 42, 160°.

All compounds gave correct N-analysis.

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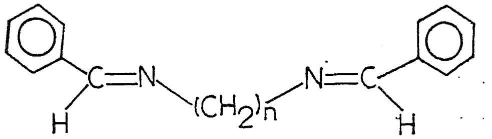
---

1. McKenzie, A. and Bate, S. C., *J. Chem. Soc.*, 1915, **197**, 1681.
2. Sandhu, J. S., Sethi, P. S. and Suresh Mohan, *J. Ind. Chem. Soc.*, 1971, **48**, 89.

MIXED LIGAND COMPLEXES OF COBALT (III) CONTAINING N, N′-TETRAMETHYLENEBIS (SALICYLIDENEIMINE) AND BIDENTATE LIGANDS

In recent years there has been considerable interest on the transition metal complexes of quadridentate Schiff bases I[1-4]. As the number of the carbon atoms in the methylene bridge increases from $n = 2$ to $n = 3$ or 4 the stereochemistry of the metal (II) complexes [where M = Co (II) and Cu (II)] changes from square planar to tetrahedral[5-7]. We have recently reported the synthesis and characterisation of cobalt (III) complexes of saltn[8], where saltn = N, N′-trimethylenebis (salicyiideneimine). In this preliminary communication we report the syntheses of several new cobalt (III) mixed ligand complexes of salbn and bidentate ligands like acetylacetone, acetoacetanilide, tropolone, N-benzoylphenylhydroxylamine and picolinic acid. The complexes have been characterised by elemental analysis, electronic spectra, conductance and magnetic measurements.



I. n = 2, salen
n = 3, saltn
n = 4, salbn

Co (salbn), where salbn = N, N′-tetramethylenebis (salicylideneimine), was prepared according to the method of Hariharan and Urbach[5]. [Co (salbn)]$_2$O$_2$ was prepared wy following a similar procedure as described[9] for the synthesis of [Co (salen)]$_2$O$_2$, where salen, = N, N′-ethylenebis (salicylideneimine). The mixed ligand complexes were prepared by reacting [Co (salbn)]$_2$O$_2$ (0·0014 mole) and the appropriate bidentate ligand (0·0028 mole) in boiling ethanol. The resulting green solution was filtered and the partial evaporation of the filtrate gave green precipitates of the complexes. These were purified by recrystallisation from 1 : 1 mixture of chloroform and ethanol. The analytical and electronic spectral data of the complexes are given in Table I.

TABLE I

Analytical and electronic spectral data of mixed ligand cobalt (III) *complexes*[a b]

Complex		% Co	% N	Band I cm^{-1}	Band II cm^{-1}	Band III cm^{-1}
[Co (acac) (salbn)]	Found:	12·6	6·3	16,840 (2·65)	25,640 (3·90)	38,830 (4·86)
Co $C_{23}H_{25}O_4N_2$	Reqd.:	13·05	6·19			
[Co (acan) (salbn)]	Found:	11·1	7·7	16,670 (2·51)	25,840 (3·88)	39,220 (4·82)
Co $C_{28}H_{28}O_4N_3$	Reqd.:	11·15	7·94			
[Co (tropolone) (salbn)]	Found:	12·5	5·8	16,840 (2·57)	25,970 (4·12)	39,530 (4·88)
Co $C_{25}H_{23}O_4N_2$	Reqd.:	12·45	5·91			
[Co (BPHA) (salbn)]	Found:	10·0	7·6	17,030 (2·46)	25,910 (3·78)	38,760 (4·70)
Co $C_{31}H_{28}O_4N_3$	Reqd.:	10·44	7·43			
[Co (pic) (salbn)]	Found:	12·8	9·2	17,010 (2·66)	25,840 (3·81)	38,020 (4·71)
Co $C_{24}H_{22}O_4N_3$	Reqd.:	12·42	8·84			

[a] Abbreviations: acac = deprotonated anion of acetylacetone; acan = deprotonated anion of acetoacetanilide; BPHA = deprotonated anion of N-benzoylphenylhydroxylamine and pic = deprotonated anion of picolinic acid.

[b] Figures in the parentheses indicate log ε.

The magnetic susceptibility measurements (27° C) of the complexes by the Gouy method indicate that the complexes are diamagnetic suggesting octahedral structure for these cobalt (III) complexes. The low molar conductance values of the complexes ($\Lambda M = \sim 5$ ohm^{-1} cm^2 mole^{-1}) in methanol are indicative of the non-electrolytic nature of the complexes. The complexes exhibit two ligand field bands in chloroform solution at around 17000 and 26000 cm^{-1} due to the probable transitions $^1A_{1g} \rightarrow {}^1T_{1g}$ and $^1A_{1g} \rightarrow {}^1T_{2g}$ respectively[10]. The high molar absortivity of the band at $\sim$ 26000 cm^{-1} indicates some contribution from the metal-ligand charge transfer transitions. The band at around 39000 cm^{-1} is assigned to the charge transfer and intraligand transitions. In these mixed ligand complexes the quadridentate ligand salbn adopts an unusual non-planar twisted conformation and the bidentate ligand occupies two *cis* positions similar to that of *cis-β*-[Co (acac) (salen)][11].

The synthesis of mixed ligand complexes using other bidentate OO, NO and NN donor ligands and also the synthesis of similar heterochelates with the ligands I (n = 5 and 6) are in progress, and the details will be reported in due course.

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---

1. Holm, R. H., Everett, Jr., G. W. and Chakravorty, A., *Prog. Inorg. Chem.*, 1966, 7, 83.
2. Sinn, E. and Harris, C. M., *Coord. Chem. Rev.*, 1969, 4, 391.
3. Hobday, M. D. and Smith, T. D., *Ibid.*, 1972, 9, 311.
4. Calligaris, M., Nardin, G. and Randaccio, L., *Ibid.*, 1972, 7, 385.
5. Hariharan, M. and Urbach, F. L., *Inorg. Chem.*, 1969, 8, 556.
6. Hoyt, W. C. and Everett, Jr., G. W., *Ibid.*, 1969, 8, 2013.
7. Batley, G. E. and Graddon, D. P., *Aust. J. Chem.*, 1967, 20, 885.
8. Syamal, A. and Ghanekar, V. D., *J. Coord. Chem.*, 1975, 5, 39.
9. Cozens, R. J. and Murray, K. S., *Aust. J. Chem.*, 1972, 25, 911.
10. Cotton, F. A. and Wilkinson, G., *Advanced Inorganic Chemistry*, Wiley Eastern Pvt. Ltd., 2nd Edition, 1966, p. 875.
11. Calligaris, M., Manzini, G., Nardin, G. and Randaccio, L., *J. Chem. Soc., Dalton*, 1972, p. 543.

PROTON-LIGAND FORMATION CONSTANTS AND FORMATION CONSTANTS OF Tl(I)-3:5-DINITRO SALICYLATES

ALTHOUGH the co-ordination chemistry of salicylic acid is well documented, the study of its derivative, the 3:5-Dinitro salicylic acid (DNSA), remained neglected until quite recently. Metal complexes of DNSA with Be (II)[1], Cu (II)[2,3], Ni (II)[2,4,12], Co (II)[2,4,12], Mn (II)[2], Zn (II)[2], UO_2 (II)[5,6], Fe (III)[7,8,14] V (IV)[9], Mg (II)[10], Al (III)[11], Ga (III)[11] and In (III)[11] are reported in the literature. No work seems to have been made to study the complexing tendency of DNSA with Tl (I). The present work deals with the same. The proton-ligand formation constants of DNSA and formation constants of its complexes with Tl (I) have been determined employing half-integral (HI), point-wise calculation (PC) and linear plot (LP) methods. Refined values of the formation constants have been obtained by the method of least-squares.

Experimental

All the chemicals used were of Analar grade.

Polymetron model CL-41 pH-meter was used for pH-metric titrations conducted at 30° C. Measurements were made with an accuracy of ± 0·05 pH units and the reproducibility of the readings was of the same order.

pH-metric titrations of solutions of (i) free $HClO_4$, (ii) free $HClO_4$ + DNSA and (iii) free $HClO_4$ + DNSA + Tl(I), were performed in 50% (v/v) aqueous ethanol medium against standard NaOH solution while maintaining the ionic strength at 0·1 M $NaClO_4$.

The experimental set up and method of calculation is the same as described earlier[15–17].

TABLE I

Proton-ligand formation constants of DNSA

pH	$\bar{n}_A$	$\log \frac{\bar{n}_A - 1}{2 - \bar{n}_A}$	pH	$\bar{n}_A$	$\log \frac{\bar{n}_A}{1 - \bar{n}_A}$
2·1	1·47	−0·052	7·6	0·83	+0·688
2·2	1·36	−0·249	7·8	0·72	+0·412
2·4	1·23	−0·524	8·0	0·59	+0·158
2·8	1·10	−0·954	8·2	0·44	−0·092
3·0	1·08	−1·060	8·4	0·38	−0·212
3·2	1·05	−1·278	8·6	0·29	−0·380
3·4	1·05	−1·278	8·8	0·22	−0·720

Results and Discussion

The Irving-Rossotti expression[18] was used for the calculation of $\bar{n}_A$, $\bar{n}$ and p(L). In the calculation of $\bar{n}_A$, $\bar{n}$ and p(L), the concentrations were corrected for the changes in volume as a result of addition of the alkali during the titrations. A series of values of $\bar{n}_A$, $\bar{n}$ and p(L) corresponding to different B-values (pH-meter readings) were calculated (Tables I–II) and the formation curves for the ligand and the complex were obtained by plotting $\bar{n}$ *vs.* pH and $\bar{n}_A$ vs. p(L) respectively (Figs. 1–2). The values of log pK_1^H, log

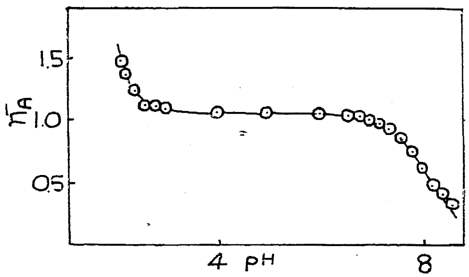

FIG. 1

TABLE II

Formation constants of Tl (I)– DNSA

pH	$\bar{n}$	$p(L)$	$\log \frac{\bar{n}}{1-\bar{n}}$	$\bar{n}_A$	$\bar{n}$	$p(L)$	$\log \frac{\bar{n}-1}{2-\bar{n}}$
1·048	0·429	6·400	−0·123	1·036	1·125	4·989	−0·845
1·048	0·573	6·167	+0·049	1·031	1·184	4·875	−0·434
1·048	0·685	5·933	+0·339	1·028	1·286	4·710	−0·196
1·042	0·795	5·679	+0·589	1·024	1·389	4·580	+0·073
1·041	0·927	5·230	+1·107	1·021	1·542	4·418	+0·481

TABLE III

Method	log pK_1^H	log pK_2^H	log K_1	log K_2
HI	8·15	2·07	6·30	4·45
PC	8·21	2·00	6·27	4·29
LP	8·17	2·02	6·25	4·45
LS	..	..	6·01	4·35
Average value	8·17	2·03	6·21	4·38

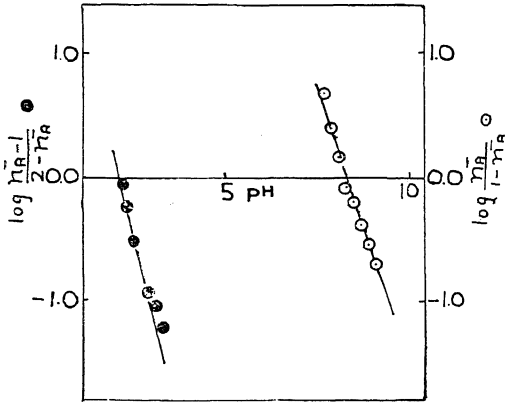

FIG. 3.

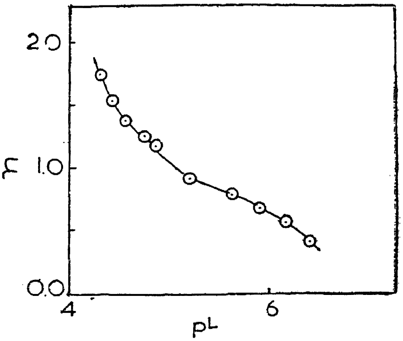

FIG. 2.

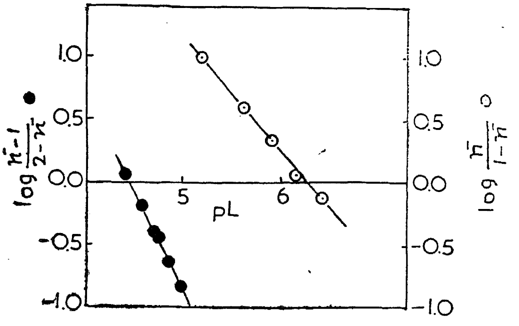

FIG. 4.

pK_{H2} and also log K_1, log K_2 were recorded directly from the respective formation curves from the integral values of $\bar{n}_A$ and $\bar{n}$ and are recorded in Table III. Refined values were then obtained by point-wise calculations as well as linear plot method (Figs. 3 4). Least-squares' method is also employed for the calculation of log K_1 and log K_2.

The values of proton-ligand formation constants of DNSA can be compared with those of Chaudhur- and Taylor[12], Dube and Dhihdsa[13] and Jahagirdar and Khanolkar[2]. Because of the different experimental conditions used in the present investigation our values are slightly different, however, the order of proton-ligand formation constants are similar.

Perusal of Table II (Fig. 2) shows that $\bar{n}$ values never exceed 1·6 indicating thereby the formation of only 1:1 and 1:2 complexes of Tl(I) with DNSA. In view of very low concentration of Tl(I) (1×10^{-4} M) used in the titration, formation of poly-nuclear complexes may be ignored. Titrations performed with different ratios of metal to ligand showed that the formation constants are independent of metal ion concentration.

The ratio log K_1/log K_2 is positive and that the separation factor between the first and the second formation constants is well within the expected range. A large difference between log K_1 and log K_2 and a high value of log K_1/log K_2 is due to the possible steric hindrance on the linking of the second ligand molecule expected for Tl(I) ion.

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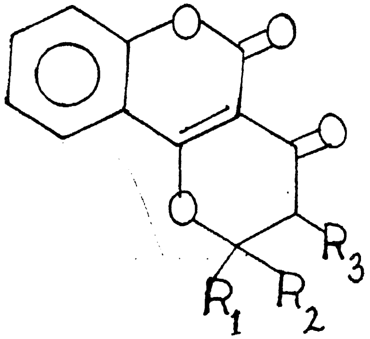
---

1. Dube, S. S. and Dhihdsa, S. S., *Z. Naturforsch*, 1969, **24**- 967.
2. Jahagirdar, D. V. and Khanolkar, D. D., *J. Indian Chem. Soc.*, 1972, **49**, 1105.
3. Vassilliadis, G., Colovos, G. and Karayiannidis, P., *Chim. Chronika*, 1966, **31**, 46.
4. Dube, S. S. and Dhihdsa, S. S., *Indian J. Chem.*, 1969, **7**, 823.
5. Bartusek, N., *Collect, Czech. Chem. Commun.*, 1967, **32**, 116.
6. Dube, S. S. and Dhihdsa, S. S., *J. Inorg. Nucl. Chem.*, 1970, **32**, 104.
7. Vassilliadis, C., Mahoussatis, G. and Colovas, G., *Chem. Chronika*, 1964, **29**. 322.
8. —., Colovas, G. and Cohstandih, D., *Ibid.*, 1966, **31**, 88.
9. Dube, S. S. and Dhihdsa, S. S., *J. Inorg. Nucl. Chem.*, 1970, **32**, 543.
10. Taylor, R. S., *Talanta*, 1974, **21**, 1210.
11. Kuznetsov, V. I. and Basavgih, N. N., *Zh. Neorg. Khin.*, 1962, **7**, 814.
12. Chaudhuri, P. and Taylor, R. S., *Anal. Chim. Acta*, 1975, **78**, 451.
13. Dube, S. S. and Dhihdsa, S. S., *Curr. Sci.*, 1968, **37**, 642.
14. Kawassiades, C. Ph. and Papavassicioh, O. Ch., *Chim. Chronika*, 1966, **31**, 67.
15. Khadikar, P. V. and Ameria R. L., *J. Indian Chem. Soc.*, 1973, **50**, 389.
16. —., Poonia N. S. and Kakkar, S. N., *Indian J. Chem.*, 1973, **11**, 709.
17. —, and Ameria R. L., *J. Indian Chem. Soc.*, 1972, **49**, 1049.
18. Irving, H. M. and Rossotti, H. S., *J. Chem. Soc.*, 1954, p. 2904.

SYNTHESIS OF CHROMANONES FROM 4-HYDROXYCOUMARIN

IN a previous communication[1] on the cyanoethylation of hydroxycoumarins, leading to the synthesis of different coumarino-chromanones, we have reported that the cyanoethylation of 4-hydroxycoumarin with acrylonitrile does not take place under different experimental conditions.

We now wish to report a one step synthesis of coumarino-chromanones from 4-hydroxycoumarin by heating the latter with acrylic, α-methylacrylic, crotonic, β, β-dimethylacrylic and cinnamic acids in the presence of PPA at about 120° for five hours. In all cases the coumarino-chromanones were isolated as pale yellow or brown crystalline solids from alcohol in yields ranging from 30 to 35%. The structures of the coumarino-chromanones were evident by the formation of crystalline 2, 4-DNPs as well as from their i.r. and n.m.r. spectral data.



I; $R_1 = R_2 = R_3 = H$
196–97°, 2, 4 D.N.P. M.P. 258–60°
II; $R_1 = R_2 = H$; $R_3 = Me$;
213–14°, 2, 4 D.N.P. M.P. 254–55°
III; $R_1 = Me$; $R_2 = R_3 = H$;
209–10°, 2, 4 D.N.P. M.P. 264–65°
IV; $R_1 = R_2 = Me$ $R_3 = H$;
184–85°, 2, 4 D.N.P. M.P. 249–51°
V; $R_1 = Ph$; $R_2 = R_3 = H$;
258–60°,

Compound V was crystallised from formic acid. The i.r. (nujol) spectrum of the coumarino-chromanones showed two bands around 1725 and 1680 cm^{-1} corresponding to the C = O band of the coumarin and the chromanone respectively. The n.m.r. ($CDCl_3$ + DMSO) of I, II and III showed the following signals (I): δ2·9 (2H; *t*; CH_2 next to C = O; J = 3Hz); δ 4·95 (2H; *t*; CH_2 next to O; J = 3Hz); δ 7∼8 (4H; *m*; aromatic); II δ 1·2 (3H; *d*; CH_3; J = 3·5 Hz); δ 2·8 (1H; *m*; >CH); δ 4·6–4·8 (2H; *t*; CH_2, J : 3Hz); δ 7∼8 (4H; *m*; aromatic); III in CF_3COOH δ 1·8∼1·9 (3H; *d*; CH_3; J = 6Hz);

δ 3 (2H; broad S; CH$_2$); δ 5·1 ∼ 5·4 (1H; broad S; >CH); δ 7·4–8·4 (4H; *m*; aromatic).

An important evidence for the chromanone structure of the above compounds was also obtained by the fact that the compound IV prepared by the above method was found to be identical (m.m.p.; i.r.) with the chromanone synthesized by Shizuri *et al.*,[2] by the condensation of 4-hydroxycoumarin with β-β-dimethyl-acryloyl chloride in the presence of pyridine.

We have also observed that the above synthesis of coumarinochromanones is a general one and takes place with 6-, 7- and 8-methyl 4-hydroxy coumarins with the above α, β-unsaturated acids in the presence of PPA.

All the compounds gave satisfactory C, H, N analysis.

We are thankful to Ciba-Giegy Research Centre and Hoechst Research Centre for obliging us with i.r. and n.m.r.

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---

1. Merchant, J. R. and Patell, J. R., *J. Chem. Soc.* 1969, p. 1545.
2. Shizuri, Y., Kato, K., Hirata, Y. and Yamamura, S., *J. Chem. Soc.*, 1969, p. 2774.

BIS (2-METHYL-$\Delta^{2(3)}$-OXADIAZINE-4, 6-DIONE)-5-SULFONYL

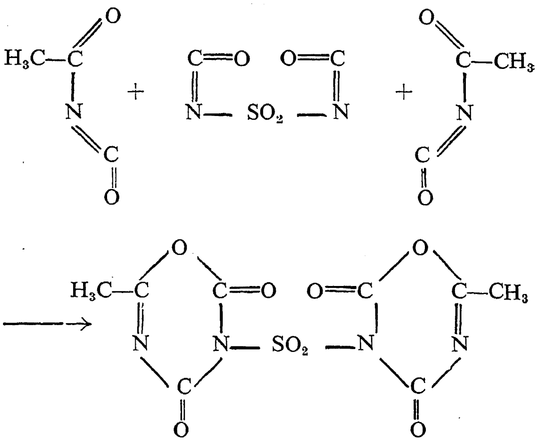
THE reaction of acetylisocyanate with sulfonyldiisocyanate leads to bis (2-methyl-$\Delta^{2(3)}$-oxadiazine-4, 6-dione)-5-sulfonyl.

Ulrich described the reaction of acetylthiocyanate with methylisocyanate[1]. The preparation of bis (2-methyl-$\Delta^{2(3)}$-oxadiazine-4, 6-dione)-5-sulfonyl, by the reaction of acetylisocyanate and sulfonyldiisocyanate, is analogous to the previously reported preparation for 2,5-dimethyl-$\Delta^{2(3)}$-oxadiazine-4, 6-dione[2], 2-methyl-5-phenyl-$\Delta^{2(3)}$-oxadiazine-4, 6-dione[3] and 3-chloro-sulfonyl-6-methyl-2H-1, 3, 5-oxadiazine-2, 4 (3H)-dione[5]. The reaction was complete in $\frac{1}{2}$ hour, at 60°–70° C, yielding a brown compound (I) (decomp. at 82° C).

The ^{1}H–NMR spectrum of bis (2-methyl-$\Delta^{2(3)}$-oxadiazine-4, 6-dione)-5-sulfonyl, which was recorded in deuterium dimethylsulfoxide, shows a signal at $\tau = 7 \cdot 9$ ppm for the methyl protons.

The I.R. spectrum (in KBr) shows the usual absorptions for the C = O, S = O and C–O–C groups at 1710, 1175 and 1050 cm^{-1}, respectively. The mass spectrum was difficult to interpret due to thermal decomposition in the inlet system.

Reaction Scheme:



(I)

EXPERIMENTAL DATA

1. *Preparation of acetylisocyanate* (*see* ref. 5).
2. *Preparation of $C_8H_6N_4O_8S$ (I).*—Acetylisocyanate (2·9 g.; 0·033 mole) was heated with sulfonyldiisocyanate (2·5 g; 0·017 mole) at 60–70° C for $\frac{1}{2}$ hour. Bis (2-methyl-$\Delta^{2(3)}$-oxadiazine-4, 6-dione)--5-sulforyl, crystallized on cooling (yield 1·2 g; 45%). (Found: C—30·07%, H—2·13%, N—17·85% and S—10·72%). (Calculated: C—30·20%, H—1·88%, N—17·53%, O—40·25% and S—10·06%).

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---

1. Ulrich, H., *Cycloaddition Reactions of Heterocumulenes*, Academic Press, 1967, Table 12, *see* also Goerdeler, J. and Schenk, H., *Chem. Ber.*, 1965, **98**, 2954 and 3831.
2. Singh, N. and Latscha, H. P., *Z. Naturforsch.*, 1970, **25B**, 1180.
3. — and —, *Curr. Sci.*, 1972, **41**, 176.
4. Sridhara, Narendra, S. and Latscha, H. P., *Z. Naturforsch.*, 1975, **30B**, (In press).
5. Billeter, O. C., *Ber. dtsch. chem. Ges.*, 1903, **36**, 3213.

TRILETE SPORES FROM KALADGI BASIN, SOUTH INDIA

PRESENT WORK

DURING the course of our micropaleophytological studies of the Kaladgi group of rocks, certain interesting trilete spores belonging to Group Triletes are recognised for the first time. Samples containing these spores are recovered from 'Chitrabhanukot Dolomite' and 'Chikshellikere Limestone' of the Lokapur Formation and from 'Machkandi Argillite' of Mudhol Formation—a new classification established for the Kaladgi Group (Viswanathiah, 1974—under publication).

BADAMI GROUP

Unconformity

KALADGI GROUP	MUDHOL FORMATION	Machkandi argillite Laksanhatti dolomite Niralkeri breccia Vajramatti quartz-arenite.
	LOKAPUR FORMATION	Yadhatti argillite Petlur limestone Jalikatti argillite Chikshellikeri limestone Chitrabhanukot dolomite.
	BAGALKOT FORMATION	Manoli argillite Mahakut breccia Bilgi quartz-arenite Salagundi conglomerate

—Unconformity—

DHARWAR SUPER GROUP

Occurrence of trilete spores are reported from Vendian and Cambrian deposits in different parts of U.S.S.R. (Timofeev, 1960, 1966). Trilete spores much different from those described in this paper have been recorded from Dharwars (Venkatachala *et al.*, 1973), Bhimas (Venkatachala and Rawat, 1973) and Vindhyans (Salujha *et al.*, 1971). The trilete mark in the spore from the Dharwar material is very faint and only a tetragonal compression mark. The mark observed in the specimens of the present study is distinct and prominently developed wherein each ray extends from 1/3 radius to the end of periphery. Detailed morphology and dimensions presumably bear phylogenetic relationship with similar spores or spore-like bodies of the early Precambrians on one hand and with those occurring from Cambrian onwards on the other.

Taxonomic notes :

Group: TRILETES Reinch, 1881.
Sub-Group: OLIGOTRILETES TIMOFEEV, 1963
Genus: *Leioligotriletum* Timofeev, 1958.
Type species: *Lieioligotriletum crassum* (Naumova) Timofeev, (1949) 1959 (Fig. 1)

Description: Spore body pale brown, spheroidal, 13 × 12 μ; wall 1 μ thick, single layered, smooth surface opaque; triradiate slit distinct, peripheral rays short, of the three rays two are more prominent and extend for 2 μ, 1 μ thick, third ray extend for 1·5 μ.

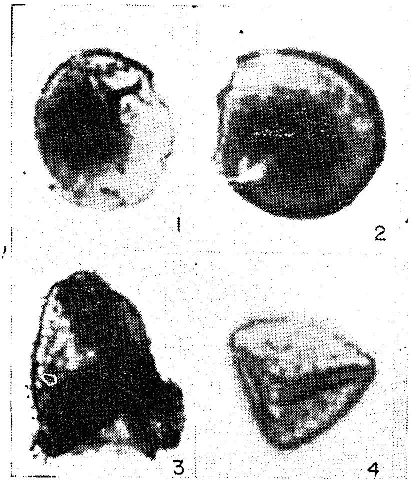
Leioligotrilelum sp. Fig. 2.

Description: Spore body brown, spherical, 14 μ; wall 1 μ thick, smooth, surface opaque; trilete rays distinct, short, two rays are prominent, extend till the periphery.

Genus: *Stenozonoligotriletes* Timofeev, 1958.
Type species: *Stenozonoligotriletes validus* Timofeev, 1958.

Stenozonoligotriletes sp. (Fig. 4)

Description: Sub-triangular (equilateral), spore body brown, 9 μ; wall 1 μ thick, punctate, surface subopaque; trilete slit broad and distinct, rays extend almost upto the periphery, one of the rays is longer, and extends for 6·5 μ, 1·5 μ wide.



(Magnification × 500)

FIGS. 1–4. Fig. 1. *Leioligotriletum crassum* (Naumova) Timofeev. Fig. 2. *Leioligotriletu* sp. Fig. 3. *Archaeozonotriletes* sp. Fig. 4. *Stenozonoligotriletes* sp.

Sub-Group: LOPHOZONOTRILETES Naumova, 1937

Genus: *Archaeozonotriletes* Naumova, 1953.

Archaeozonotriletes sp. (Fig. 3)

Description: Spore body brown, sub-triangular, 38 × 28 μ; wall 1 μ thick, smooth, slightly wavy, surface subopaque; trilete rays distinct, enclosed in thick folds, rays extend upto the periphery, folds 10 μ wide.

The Junior author (A. P. Mahalakshmamma) acknowledges the help extended by Sri. M. J. Chandrasekhara Gowda, Department of Geology, Manasa Gangotri, by providing the required material for this work.

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Mysore-6, *October* 6, 1975.

---

1. Salujha, S. K., Rehman, K. and Rawat, M. S., *Rev. Palaeobot. Palynol.*, 1971, **11**, 65.
2. Tschibrikova, E. V., *Akad. Sci. USSR*, Publications Sciences, Moscow, 1972.
3. Timofeev, B. V., *Geol. Foren. Forhandl.*, 1960, **82** (1), 29.
4. —, Micropaleophytological research into ancient strata. *Sciences*, 1966, Moscow, 147 pp., 189 plates.
5. Venkatachala, B. S., Bhandari, L. L., Chaube, A. N. and Rawat, M. S., *Paleobot.*, 1973, **21** (1).
6. — and Rawat, M. S., *Geophytol.*, 1973, **2** (2), 107.
7. Viswanathiah, M. N., *Lithostratigraphy and Structure of the Kaladgi Group, Karnataka State*, 1974. (Under publication).

NOTE ON THE PRESENCE OF TRIASSIC AND PERMIAN SEDIMENTS IN SUB-SURFACE, SHUMARWALI TALAI AREA. JAISALMER DISTRICT, RAJASTHAN

THE Pre-Jurassic history of Western Rajasthan shelf is little known. So far, the presence of Triassic and Permian sediments in sub-surface have not been reported from this area.

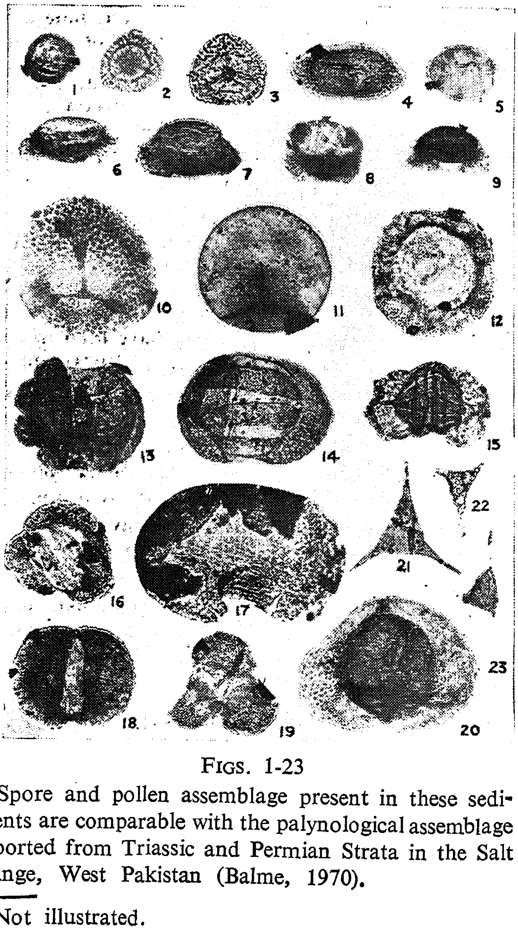
The objective of this note is to report the first discovery of the presence of sub-surface Triassic and Permian sediments in the Shumarwali Talai area of Rajasthan. This finding further clears the doubt on the presence of Permo-Triassic sediments in Western Rajasthan.

In the course of exploration for hydrocarbons in one of the exploratory well at Shumarwali Talai (27° 11′ 12″: 70° 17′) below the Lathi formation (Liassic, Lukose, 1971), about 515 metres thickness of sediments have been penetrated. On the basis of well preserved assemblage of spore and pollen present in these sediments, 260 metres of sediments are of Triassic age, and below this succession, 255 metres sediments are of Permian age (full thickness of Permian not penetrated).

Lithologically the sediments of Triassic age comprise mainly of greyish white, occasionally dull brown, medium to coarse grained, quartzose, angular to subangular, non-calcareous, sand and soft to moderately hard, non-calcareous claystone. The palynological evidence suggests that these sediments have been deposited under non-marine environment. The characteristic spore and pollen assemblage recorded in the Triassic sediments comprises of *Gliscopollis* sp. (Fig. 1), *Staplinisporites* sp. (Fig. 2), *Tigrisporites* sp. (Fig. 3), *Regulatisporites* sp. (*), *Guttatisporites* sp. (*), *Lundbladispora* sp. (*), *Bascanisporites* sp. (*), *Ovalipollis* sp. (Figs. 4 and 5), *Taeniaesporites* sp. (Fig.6), *Striatites*. sp. (Fig. 7), *Distriatites* sp. (Fig. 8), *Samaropollenites* sp. (Fig. 9).

Underlying the above succession, the sediments of Permian age comprise of mainly grey, brownish white, medium to coarse grained, poorly sorted, non-calcareous, quartzose, occasionally pyritic sand and variegated soft to moderately hard claystone. Subordinate percentage of carbonaceous grey shale is also present. The palynological evidence suggests these sediments have been deposited under shallow marine environment.

The characteristic spore and pollen assemblage recorded in the Permian sediments comprises of—*Cyclobacutatisporites* sp. (Fig. 10), *Calmospora* sp. (Fig. 11), *Verrucosisporites* sp.(*), *Sulcatisporites* sp.(*)., *Virkkipollenites* sp. (Fig. 12), *Guttulapollenites* sp. (Fig. 13), *Corrisacites* sp. (Fig. 14), *Hamia-pollenites* sp. (Fig. 15), *Mahudapoltenites* (Fig. 16), *Distriatites* sp, (Fig. 17), *Protohaploxypinus* sp. (*), *Striatopodocarpites* (*), *Falcisporites* sp. (Fig. 18), *Trochosporites* sp. (Fig. 19), *Potonisporites* (Fig. 20), *Lueckisoporites* spp. (*), *Illinites* sp. (*), *Micrhystridium* spp. (*), *Veryhachium valensii* (Fig. 21), *Veryhachium* sp. (Figs. 22–23).



FIGS. 1-23

Spore and pollen assemblage present in these sediments are comparable with the palynological assemblage reported from Triassic and Permian Strata in the Salt Range, West Pakistan (Balme, 1970).

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* Not illustrated.

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1. Balme, B. E., *Univ. Kansas, Dep. Geol. Spec. Publs.*, 1970, p. 305
2. Lukose, N. G., *Proc. Seminar Paleopalynology, Indian Stratigraphy*, 1971, p. 155

CHANGES IN PHOSPHOLIPID CONTENT OF BLOOD AND MALPIGHIAN TUBULES OF *EPILACHNA DODECA-STIGMA* MULS. (COLEOPTERA)

Although numerous analyses of phospholipids of vertebrates have been made, similar studies on insects are relatively recent[1-7]. Except for the works of Crone (1964)[5], Taylor and Hodgson (1964)[6], Bridges and Watts (1975)[7] others are on phospholipid composition of whole insects. Similarly much emphasis is given to the influence of diets on the patterns and composition of phospholipids in insects[8-10]. It is reasonable to believe that the concentration of phospholipid in individual organs might be different from that of the whole organism. So the present study was undertaken to investigate the changes in the concentration of phospholipid in the blood and malpighian tubules of *Epilachna dodeca-stigma* under various physiological conditions.

Male *E. dodeca-stigma* of the same age and more or less of the same size were starved for two days in separate jars each containing 8–10 insects. They were then fed regularly with the flowers of *Hibiscus rosachinensis* for 8 days. They were later transferred to separate jars and kept without food along with a control set of insects that had free access to these flowers. Blood and malpighian tubules were collected at regular intervals employing adequate number of insects. Total phospholipid was estimated by the method of Annino (1964)[11].

The analytical data of the total phospholipid content in blood and malpighian tubules of normally fed and starved *E. dodeca-stigma* are shown in Table I. The

Table I

Phospholipid in ♂ *E. dodeca-stigma*

Period	Malpighian tubules mg/100 mg		Blood mg/100 ml	
	Fed	Starved	Fed	Starved
0 hour	0·39 ± 0·002	0·41 ± 0·001	15·4 ± 0·04	14·8 ± 0·16
8 hours	0·45 ± 0·001	0·43 ± 0·002	16·2 ± 0·18	16·4 ± 0·81
16 hours	0·40 ± 0·001	0·43 ± 0·008	15·1 ± 0·21	13·2 ± 0·21
24 hours	0·40 ± 0·002	0·45 ± 0·002	14·6 ± 0·08	16·2 ± 0·64
32 hours	0·40 ± 0·006	0·34 ± 0·008	15·8 ± 0·3	14·6 ± 0·32
40 hours	0·41 ± 0·01	0·32 ± 0·006	14·8 ± 0·18	14·3 ± 0·01
48 hours	0·39 ± 0·002	0·34 ± 0·004	15·1 ± 0·13	13·2 ± 0·82
56 hours	0·40 ± 0·001	0·32 ± 0·008	15·3 ± 0·14	10·8 ± 0·81
64 hours	0·42 ± 0·002	0·33 ± 0·006	14·8 ± 0·41	9·1 ± 0·41
72 hours	0·41 ± 0·004	0·29 ± 0·004	15·2 ± 0·62	7·4 ± 0·62
80 hours	0·40 ± 0·008	0·26 ± 0·001	15·3 ± 0·38	5·2 ± 0·56
88 hours	0·39 ± 0·001	0·24 ± 0·008	15·1 ± 0·81	4·9 ± 0·82
96 hours	0·41 ± 0·006	0·21 ± 0·004	14·8 ± 0·64	3·6 ± 0·91

phospholipid content ranges from 0·39 ± 0·002–0·45 ± 0·001 mg/100 mg of malpighian tubules and 14·6 ± 0·008–16·2 ± 0·18 mg/100 ml of blood. The total phospholipid content of these tissues considerably decreases during starvation. However after a full meal both these tissues acquire a slightly elevated level (Table I). The blood phospholipid, with an initial decline at 16 hours starvation depicts a slight increase till 24 hours, thereafter, it gradually decreases till 96 hours. The malpighian tubules on the other hand, with an initial marked increase in phospholipid content upto 24 hours, show a sharp decline at 32 hours whereafter maintaining more or less a constant level upto 64 hours, once again exhibit a gradual fall till 96 hours.

Since the insects die or become inactive at the end of fourth day of starvation, no estimation could be made beyond this period. The fall in the blood phospholipid content is more significant than that in the malpighian tubules. Though information on the tissue lipids of other insects are not available except for silkworm[12], it is worth mentioning that phospholipid content of blood and malpighian tubules of normally fed male *E. dodeca-stigma* is more or less constant.

The author wishes to express his thanks to Dr. K. J. Joseph, Professor and Head of the Department, for his encouragement.

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1. Fast, P. G., *Mem. ent. Soc. Con.*, 1964, **37**, 1.
2. Kamienski, F. X., Newburgh, R. W. and Brooks, V. T., *J. Insect Physiol.*, 1965, **11**, 1533.
3. Khan, M. A. and Hodgson, E., *Ibid.*, 1967, **13**, 653.
4. Yadava, R. P. S. and Musgrave, A. J., *Comp. Biochem. Physiol.*, 1972, **41B**, 425.
5. Crone, H. D., *J. Insect Physiol.*, 1964, **10**, 499.
6. Taylor, I. F. and Hodgson, E., *Ann. Ent. Soc. Amer.*, 1964, **57**, 795.
7. Bridges, R. G. and Watts, S. G., *J. Insect Physiol.*, 1975, **21**, 861.
8. Miller, G. J. and Blankenship, J. W., *Ibid.*, 1973, **19**, 65.
9. Dwivedy, A. K. and Bridges, R. G., *Ibid.*, 1973, **19**, 559.
10. Castillon, M. P., Catalan, R. E., Jimenez, C., Madariaga, M. A., Muncio, A. M. and Suarez, A., *Ibid.*, 1974, **20**, 507.
11. Annino, J. S., *Clinical Chemistry: Principles and Procedures*, Published by J and A Churchill Ltd., London, 1964, p. 272.
12. Shridhara, S. and Bhatt, J. V., *J. Insect Physiol.*, 1965, **11**, 499.

A SIMPLE NEW TECHNIQUE TO DEMONSTRATE THE PRESENCE OF GIANT NERVE CELLS (RETZIUS' CELLS) IN THE SUB-PHARYNGEAL AND SEGMENTAL GANGLIA OF THE COMMON INDIAN CATTLE LEECH, *POECILOBDELLA GRANULOSA*

CENTRAL nervous system of leeches consists of a pair of cerebral ganglia, a pair of peri-pharyngeal connectives, a sub-pharyngeal ganglionic mass, composed of four pairs of ganglia fused together, and a ventral nerve-cord, made up of segmental ganglia. Although a lot of work has been done on the Indian leeches[1], even the presence of giant nerve cells (Retzius' cells) has somehow completely escaped the attention of all the workers. A very simple technique has been developed to demonstrate these cells. Chilled C_α–formol – fixed frozen sections (5–10 μ) of the (freshly taken out) central nervous system of leeches were suitably processed for alkaline phosphatase by the method of Gomori[9]. These paired cells can be seen clearly embedded in the central neurofibrillar network of the sub-pharyngeal and segmental ganglia and depicting extremely intense enzymatic localization. The activity is in extreme contrast to the characteristic punctate reaction in the surrounding neurofibrils, but similar to the one seen in the neurons, surrounding the core. Some work has been done on the gross morphology, neuronal geometry and electrophysiology of the Retzius' cells[3-6,10]. However, their precise function is not yet known. Considering that each cell is electrically coupled with its contralateral partner, but not with the partners in the adjacent ganglia, paucity of their fine arborizations and thereby minimal synaptic contacts, their involvement in the control of dominating common motor responses and the stereo-typed behaviour of the leeches has been speculated.

The demonstration of such giant cells by the technique, communicated here, presents a fascinating biological material for neuro-anatomical and physiological work. It can also be used in laying to rest the metameric controversies in certain leeches. There is a significant segmental telescoping in leeches owing to cephalization. This, as well as the absence of inter-segmental septa in adult leeches put the criterion of segmental demarcation at stake. There have been two schools of thought on this issue. Some workers emphasize that the first annulus, which bears the characteristic tactile and photoreceptor sensillae, should be regarded as the external evidence of the beginning of a segment[1,7]. Others[2,8] regard the nervous system as the basis for determining the segmental limits, a view widely held now. Such an idea, however, requires a thorough study of the ganglional geometry and pattern of innervation before a final word about the segmental condition in a leech can be said. The present method of studying the Retzius' cells provides a short cut to this neuronal method of determining segmental constitution or composition. A glance at a section, processed accordingly, gives a clear-cut idea about the number of segments involved in the sub-pharyngeal ganglional architecture, which is otherwise very complex due to cephalization, segmental telescoping and lack of internal septa in adult leeches.

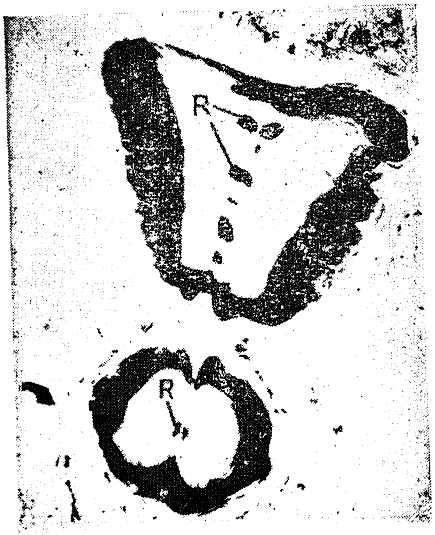

FIG. 1. Horizontal section of the sub-pharyngeal ganglionic mass along with the first pair of segmental ganglia, showing strong activity in the Retzius' cell (R) and their connecting fibres (IF), × 39.

Thanks are due to Prof. H. B. Tewari, for encouragements and to I.C.M.R. for providing fellowship to one of us (G. C. Mishra).

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1. Bhatia, M. L., *Hirudinaria. The Indian Zool. Memoirs* (Bahl. K. N., Ed.), Lucknow Publishing House, Lucknow, 1941, 8.
2. Castle, W. E., *Proc. Am. Acad. Art. Sci.*, 1900, 35, 285.
3. Coggeshall, R. E., *Anat. Rec.*, 1972, 172, 489.
4. Eckert, R., *J. Gen. Physiol.*, 1963, 46, 573.
5. Lent, C. M., *Comp. Biochem. Physiol.*, 1972, 42 A, 857.
6. —, *Ibid.*, 1973, 44 A, 35.
7. Miller-Miller, J. A., *Ohio, J. Sci.*, 1945, 45, 233.
8. Moore, J. P., *Proc. Ac. Nat. Sci., Philad*, 1900, 5, 50.
9. Pearse, A. G. E., *Histochemistry : Theoretical and Applied*, J. and A. Churchill, Ltd., London, 1968.
10. Retzius, G., *Biologische Untersuchungen Nene Folge II*, Sampson and Wallin, Ochholm, 1891.

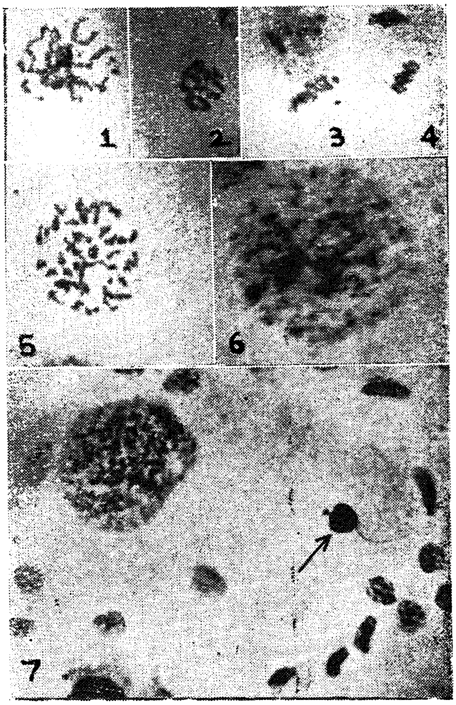
OCCURRENCE OF ENDOPOLYPLOIDY IN ABDOMINAL SCENT GLANDS OF *CHRYSOCORIS PURPUREUS* (WESTW.) PENTATOMIDAE—HETEROPTERA

REVIEW of the literature shows that there is a paucity of knowledge on the differentiation and nuclear changes undergone by the first pair of abdominal scent glands of *Chrysocoris purpureus*. However, scent glands in certain other Heteroptera and their role in defence or pheromone secreting activity have been well demonstrated[1]. The first pair of abdominal scent glands in *C. purpureus* are of developing non-functional type in all the instars whereas the 2nd and 3rd pairs are fully functional in all the nymphs. After the final instar the 2nd and 3rd pairs of nymphal glands degenerate and the first pair of developing nymphal glands become functional in 2–3 days old adult bugs of both sexes. The present study was undertaken to investigate the occurrence of endopolyploid nuclei in the first pair of abdominal scent glands of adults and nymphs of both sexes of a pentatomid bug, *C. purpureus*.

In adult bugs, the abdominal scent glands are situated dorsolaterally just above the third abdominal air sac. The abdominal scent glands were dissected out and fixed in Carnoy's solution. The fixed glands were hydrated and hydrolysed in N HCl at 60° C for 8 min, and stained in feulgen solution for 2 hours. Some glands were also stained by the haematoxylin technique. The endopolyploid gland cells can easily be found in squash preparations due to their enormous size.

Post-embryonic developmental study of the gland cells clearly exhibits mitotic stages with diploid number (Figs. 1–4 ; $n = 6 + XY$)[2,3]. In the differentiating cells of the gland, endopolyploidy regularly succeeds mitosis. The nuclear division was usually not observed after third instar stage. Different degrees of ploidy were also encountered during post-embryonic development (Figs. 5 and 6). Abdominal scent gland of the adult is characterised by the enlarged secretory cell (Fig. 7), has a relatively clear boundary with characteristic giant nucleus. Each secretory cell has an exocuticular complex end-apparatus with an elongated canaliculus to drain out the cellular secretion into a reservoir[4].

The giant nuclei of both sexes exhibit different structures, either the chromatin is uniformly distributed or it is organised in the form of chromatin bundles. However, it is not possible to make accurate counts or descriptions of the chromosomes. Further, cytophotometric (*i.e.*, DNA content) measurements are essential to enunciate the degree of ploidy. A change in the gene activity may be the cause for the abrupt transition from the mitotic to the endomitotic cycle during development. The continuous activity of the chromosomes appears to be one of the advantages of endomitotic growth as it permits high synthesis of nucleic acids. Ecdysone and juvenile hormones of insects were believed to cause gene activation[5,6]. Probably, the synthetically active cells of the abdominal scent glands require an enlargement of nuclear surface.



FIGS. 1–7. Abdominal scent gland squashes of *C. purpureus*. Figs. 1–4. Mitotic stages from 3rd instar showing Pro, Meta, Ana and Telophases, × *ca* 1,100. Figs. 5 and 6. Degree of ploidy during 3rd instar, note variation in nuclear size, × *ca* 1,100. Fig. 7. Adult endopolyploid cell, its giant nucleus and end-apparatus. Arrow indicates exocuticular end-apparatus. Small interstitial nuclei adjacent to cell boundary are also seen, × *ca* 950.

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---

1. Pattenden, G. and Staddon, B. W., *Ann. Ent. Soc. Amer.*, 1972, **65**, 1240.
2. Jande, S. S., *Res. Bull* (*N.S.*), Panjab Univ., 1959, **10**, 215.
*3. Muthukkaruppan, V. R., *J. Annamalai Univ.*, **22**, 1.
4. Janaiah, C., Venkat Reddy, P. and Chari, N., *First All India Symposium on Comparative Animal Physiology*, University of Marathwada, 1975 (In press).
5. Clever, U. and Karlson, P., *Expl. Cell. Res.*, 1960, **20**, 623.
6. Lezzi, M. and Gilbert, L. T., *Proc. Natn. Acad. Sci. U.S.A.*, 1969, **64**, 498.

* Not consulted in original.

ON MONOGENETIC TREMATODES FROM *UPENEUS VITTATUS* (FORSKAL) AND *UPENEUS SULPHUREUS* (CUVIER) OF WALTAIR COAST, BAY OF BENGAL

THE recovery from the gills of the Waltair coast fishes *Upeneus vittatus* and *U. sulphureus* of a diclidophorid monogenean which appears to be new to science has allowed us to comment on monogenea of these fishes. It is well known that considerable host specificity is displayed by monogeneans and species of *Upeneus* harbour characteristic parasites. This is the first time that a comprehensive survey of Monogenea of Waltair coast fishes has been undertaken.

In the pedunculate nature of the clamps, the specimens collected from Waltair resemble the genus *Diclidophora* (Diesing, 1850) but in the limited number of testes and genital hooks as well as in the general morphology, it comes close to the genus *Upenicola* Unnithan 1966.

Unnithan[1] described *Upenicola upeneoides* from *U. vittatus* of the South-West Coast of India. Although work has been done on Monogenea of fishes of East Coast by Chauhan[2], especially in the north-eastern parts of Bay of Bengal and Monogenea in the more southern region have been studied chiefly by Ramalingam[3], no information is available on the Monogenea of fishes in the neighbourhood of Waltair coast. In view of the fact that Mamaev[4] recently described two more species, *Upenicola digitata* and *U. capheni* from *Upeneus* spp. from the gulf of Tonkin, North Vietnam, it appeared well worth reporting on the Monogenea of Waltair coast fishes *U. vittatus* and *U. sulphureus*.

The specimens obtained by us from the fishes *U. vittatus* and *U. sulphureus* are definitely different from the genus *Upenicola*. It is proposed to erect a new genus and the name *Upenicoloides* is suggested. The new genus differs from *Upenicola* in having pedunculated clamps, 12-15 testes, 6-8 genital hooks and vitellaria extending into the posthaptor. The genus occurs on the gills of both *U. vittatus* and *U. sulphureus*. The characters of the new genus combine the features of the genera *Dicltdophora* and *Upenicola*, so as to warrant the errection of a new genus.

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Andhra University,
Waltair, *October* 27, 1975.

---

1. Unnithan, R. V., *Pacific Science*, 1966, 20, 79.
2. Chauhan, B. S., *Rec. Indian Mus.*, 1953, 2, 113.
3. Ramalingam, K., *Proc. Nat. Inst. Sci. India*, 1960, 26, 352.
4. Mamaev, Yu. L., *Issledovaniya pofaune, sistematikei biokhimii gelmintov Dal'nego Vostoka*, 1972, 11, 163.

EFFECT OF INSECTICIDES ON THE NERVE CONDUCTION IN *PERIPLANETA AMERICANA* L.

Introduction

THE insecticidal action brings about functionsl changes in the nervous system (Narahashi, 1971). Yamasaki and Ishii (1954 *a*) demonstrated that DDT poisoned roaches show convulsions and symptoms of paralysis. In the DDT poisoned insects, the spontaneous discharge in the nerve cord is increased in frequency and the synaptic transmission is facilitated (Dresden, 1949; Harlow, 1958; and Heslop and Ray, 1959). The present study was conducted to provide information on the effects of insecticides on the nerve conduction.

Materials and Methods

Since electrical potential is the only signal easily observable while the nerve is in excited state, electrophysiological techniques were used in the present work. The following experimental procedure was adopted.

The nerve cords were extracted taking care not to touch the nerve cord with hand or any metallic instrument. The extracted nerve cord was fixed in electrode box containing Hoyle's locust saline. One end of the nerve cord was connected to the output of an electronic stimulator and the other to the input of a cathode ray oscilloscope. The stimulus was fed into the nerve cord and the conductivity recorded in terms of the spike height in the oscilloscope. The stimulus was given only when the nerve was out of saline. The stimuli of different frequencies and at varying time intervals given and the effect of both was seen keeping one or the other parameter constant.

The insecticides were sprayed on the body of the insects after finding out LD_{50} for each insecticide by the usual screening tests. LD_{50} for aldrin was found to be 0·3% and for BHC 0·25% the insecticidally treated animals were dissected and nerve cords extracted in an identical manner to that of controls.

Observations

It was observed that with the increase in time duration, the spike height decreased and with the increase in frequency of stimulus, the spike height increased (Tables I and II). This increase was more in case of BHC than in aldrin treated insects. In both cases, increase was so large that at high frequencies, it was not possible to record the spike height (Table II).

CATHODE RAY OSCILLOSCOPE READINGS IN UNTREATED AND TREATED *Periplaneta americana*

TABLE I

Effect of time of stimulus on the spike height
Pulse frequency: 25 (constant)

Sl. No.	Time milli seconds	Spike height (cm)		BHC treated
		Untreated	Aldrin treated	
1.	0·2	1·1	3·0	3·8
2.	0·5	1·1	2·6	3·4
3.	1·0	1·0	2·3	3·0
4.	2·0	0·9	2·1	2·6
5.	20·0	0·8	1·9	2·4
6.	50·0	0·7	1·9	2·0

TABLE II

Effect of pulse frequency of stimulus on the spike height
Time : 0·5 milli seconds (constant)

Sl. No.	Pulse frequency	Spike height (cm)		
		Untreated	Aldrin treated	BHC treated
1.	2	0·9	2·9	3·6
2.	5	1·1	3·2	3·8
3.	10	1·3	3·6	4·0
4.	25	1·4	3·9	Unrecordable
5.	50	1·6	Unrecordable	do.
6.	100	2·0	do.	do.

Discussion

The contact insecticides usually find their entry into the body and exert a toxic action at the nerve membrane or at the synaptic functions. It has been suggested (Narahashi, 1971) that conditions of ataxia, hyperactivity and convulsions observed in insects are due to neuro-muscular actions of the insecticides.

In the case of aldrin treated insects, it was seen that by giving repetitive impulses, the action potential of the nerve cord progressively increases. In BHC treated insects, the excitability is still more. Ultimately it becomes unrecordable in both cases of treatment. BHC is a stronger poison and produces more severe convolutions leading to paralysis. Lindane which is a γ-insomer of BHC, also produces similar effects (Yamasaki and Ishii, 1945 *b*). In Lindane poisoned insects, the effect on the central nervous system dominates over that of peripheral nervous system. It has been shown that the frequency of spontaneous discharges in the nerve cord is increased significantly by lindane treatment (Harlow, 1958; Yamasaki and Ishii, 1954 *b*). Symptoms of poisoning, in the present case seem to be due to a response to the hormonal action (Roan and Hoppins, 1961) or due to the release of a toxic substance (Shakland and Kearns, 1959; Hawkins and Sternburg, 1964; Blum and Kearns, 1956; Sternburg *et al.*, 1959). Symptoms may also be attributed to the destruction of the capacity to maintain normal ionic balance [Welsh and Gordon (1947), Gordon and Welsh (1948), Mullins (1954, 1955)].

Acknowledgement

The author is grateful to Dr. S. C. Rastogi for guidance and to the Head of the Department for providing necessary facilities.

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1. Blum, M. S. and Kearns, C. W., *J. Econ. Ent*,, 1956, **49**, 862.
2. Dresden, D., *Drukkerij en Uitgeverij G. W*,, Van der Wiel and Co., Arnheim, Netherlands, 1949, p. 114.
3. Gordon, H. T. and Welsh, J. H., *J. Cell. Comp. Physiol.*, 1948, **31**, 395.
4. Harlow, P. A., *Ann. Appl. Biol.*, 1958, **46**, 55.
5. Hawkins, W. B. and Sternburg, J., *J. Econ. Ent.*, 1964, **57**, 241.
6. Heslop, J. P. and Ray, J. W., *J. Insect Physiol.*, 1959, **3**, 395,
7. Mullins, L. J., *Chem. Rev.*, 1954, **54**, 289.
8. —, *Science*, *N.Y.*, 1955, **122**, 118.
9. Narahashi, T., *Adv. in Insect Physiol.*, 1971, Vol. 8, Academic Press, London and New York.
10. Roan, C. C. and Hopkins, T. L., *A Rev. Ent.*, 1961, **6**, 333.
11. Shankland, D. L. and Kearns, C. W., *Ann. Ent. Soc. Am.*, 1959, **52**, 386.
12. Sternburg, J., Chang, S. C. and Kearns, C. W., *J. econ. Ent.*, 1959, **52**, 1070.
13. Welsh, J. H. and Gordon, H. T., *J. Cell. Comp. Physiol.*, 1947, **30**, 147.
14. Yamasaki, T. and Ishii, T., *Botyu Kagau*, 1954 *a*, **19**, .1.
15. — and —, *Ibid.*, 1954 *b*, **19**, 106.

ABNORMAL DEVELOPMENT OF EARHEADS IN RICE (*ORYZA SATIVA* L.) DUE TO THE WHITE TIP NEMATODE (*APHELENCHOIDES BESSEYI* CHRISTIE, 1942)

Infestation by the white tip nematode (*Aphelenchoides besseyi*) in rice at heading stage is known to result in twisting and shortening of apical part of boot-leaf, poor emergence of panicle and reduction in earhead length[1,2]. Spikelets present a shrivelled, bleached or chaffy appearance[3]. During October, 1975 many early maturing rice varieties at heading stage on the Farm at the Central Rice Research Institute, Cuttack, showed the above symptoms and were found to be affected by the nematode. Besides, *Bala*, *Cauvery*, *Mtu*-17 mutant-2, etc., showed twisting of flag-leaf and poor emergence of panicle. In *Jayanti*, MR 1550, etc., which were flowering, only those rachii with spikelets containing the nematodes in the active stage were elongated. (Fig. 1.1). In Ac 4156, JBS 158 and JBS 1527, etc., the glumes, lemma and palea of spikelets which contained the nematodes were out-grown as thin leaf-sheath like structures (Figs. 1.2, 1.3 and 1.4). These deformities in field infestation by this nematode prompted further inoculation studies.

Inoculations were given to blooming spikelets of JBS 1527 and Ac 4156 each with (*a*) 50 active adults of *A. besseyi* with a small drop of distilled water (*b*) their extract macerated in distilled water and (*c*) distilled water. The inoculum or water drops were placed in the vortex of lemma and palea. On the 11th day, 19 spikelets in JBS 1527 inoculated with nematodes contained 7 to 38 active nematodes and immature stages. The spikelets showed the same abnormal growth of the glumes, lemma and palea as in fields (Fig. 1.5). Only four spikelets in Ac 4156 inoculated with the aqueous extract of the nematodes showed malformation (Fig. 1.6). All the treated spikelets showed kernel development.

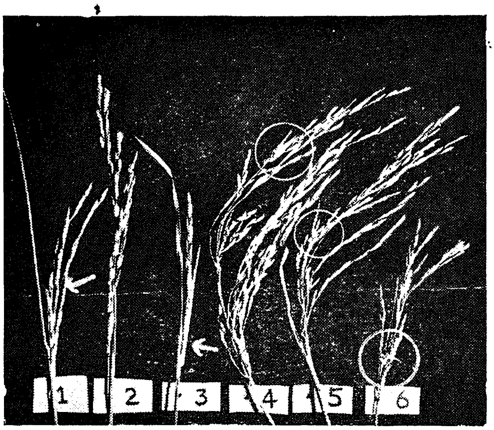

FIG. 1. Abnormal development of ears and spikelets in rice due to *A. besseyi.* (1) *Jayanti,* (2) JBS 158, (3) JBS 1527, (4) Ac 4156-field infestation. (5) JBS 1527 and (6) Ac 4156 under inoculated condition.

The above results confirmed the abnormal development of earheads due to *A. besseyi* infestation and indicated the action of a growth promoting substance. Growth stimulation of weed *Sporobolus poiertii* due to the infestation by this nematode[4] and in many dicots due to root-knot nematodes has been reported[5,6]. Abnormal growth of earheads and spikelets in rice due to *A. besseyi* may be the result of the reaction of the host tissues to the secretions of the nemaode which needs further confirmation on the exact biochemical mechanism involved in this nematode-rice plant relationship.

Central Rice Research Institute,
Cuttack-6, *November* 11, 1975.

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1. Ichinohe, M., *Japan Pesticide Information Rev.*, 1973, 14, 11.
2. Muthukrishnan, T. S., Rajendran. G. and Chandrasekharan, J., *Indian J. Nematol.*, 1974, 4, 188.
3. Haai Vuong Huu, *Instt de Res Agronomique Tropicale,* IRAT Doc, 341, Madagascar, 1972.
4. Marlett, R. B. and Perry, V. G., *Phytopathol.*, 1971, 61, 740.
5. Viglierchio, D. R., *Bot. Rev.*, 1971, 37, 1.
6. — and Yu, P. K., *Exptl. Parasitol.*, 1968, 23, 88.

LEAF FALL DISEASE OF NAGPUR ORANGES CAUSED BY *PHYTOPHTHORA NICOTIANAE* VAR. *PARASITICA* WATERHOUSE

THE occurrence of ' leaf fall ' disease of oranges (*Citrus reticulata* Blanco.) in Vidarbha (1969–70) and Nagpur (1975) is indicative of the widespread nature of this disease and its direct correlation with high humidity prevailing in July–August.

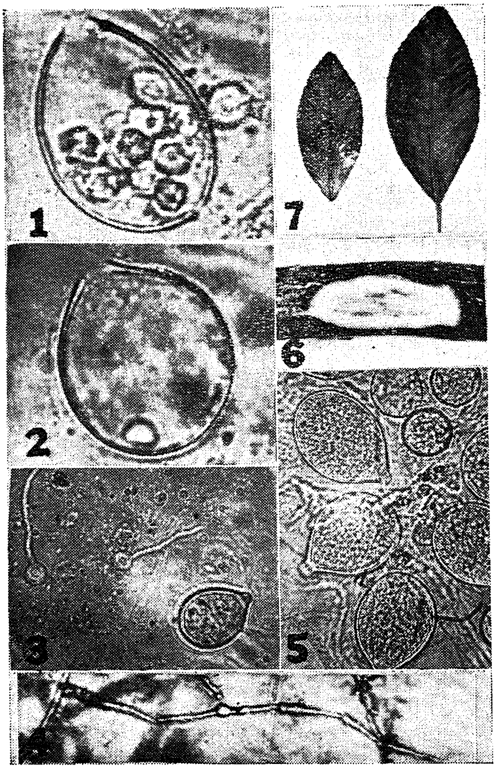
The symptoms manifest themselves as water-soaked light brown to brown patches on the leaf lamina with concentric zonation and scanty white felt of the mycelium. Tender twigs adjacent to the affected leaves and also the small berries of ' Mrugbahar ' become discoloured. Severe defoliation and berry drop leave the lower portion of the tree almost completely barren[1,6].

Investigations revealed that the fungus *Phytophthora nicotianae* var. *parasitica* Waterhouse. (Syn. *P. parasitica* Dastur.) was associated with the leaf fall syndrome. Infected leaf tissues when planted on potato dextrose agar medium yielded growth on this fungus. While the isolations from leaves yielded *P. nicotianae* var. *parasitica*, the infected tender twigs yielded in addition *Colletolrichum gloeosporioides* Penz. The infected berries yielded *C. gloeosporioides* only.

Inoculations were carried out by spraying spore suspension on (*a*) detached leaves, (*b*) fresh twigs in nutrient solution and (*c*) potted seedlings of Nagpur orange under conditions of high humidity. *P. nicotianae* var. *parasitica* alone produced typical leaf fall symptoms (Plate I, 7), while in combination with *C. gloeosporioides*, it caused twig blight symptoms. The latter singly produced only the withertip symptom (12–14 days after inoculation). In the inoculation with agar culture, discs placed on slightly injured stalk-ends of semiripe orange fruits, *P. nicotianae* var. *parasitica* alone and in combination with *C. gtoeosporioides*, produced softening and decay of fruits. The latter alone caused brown sunken lesions on the fruits (4–8 days after inoculation). Original isolates were recovered from the artificially inoculated plants and the pathogenicity confirmed. *P. nicotianae* var. *parasitica* caused softening and decay of other fruits *viz.*, *Carica papaya* L., *Solanum melongena* L., *Hibiscus esculentus* L. and *Capsicum frutescens* L. when they were inoculated as stated above. Tomatoes remained unaffected.

One-year old seedlings of eleven different root stocks were inoculated by placing discs of agar culture of *P. nicotianae* var. *parasitica* in slightly injurred bark. All root stocks except Trifoliate orange and Malta lemon developed infection within onem onth (Plate I, 6). One-month old seedlings of the above-mentioned root stocks yielded negative results when tested in soil mixed with culture of *P. nicotianae* var. *parasitica*.

The fungus *P. nicotianae* var. *parasitica* gave good growth and sporangial formation on bean extract agar, oat meal agar and papaya extract agar media. A pH of 6·0 at 27°C and 80–90% RH and continuous light favoured optimum growth of this fungus. Ultraviolet irradiation for 5 min. induced abundant sporangial formation in the culture. In hanging-drop mounts, sporangial germination by zoospore formation was optimum at 20° C (Plate I 1, 2, 3) while above 24° C, the germination by germ tube formation was more predominant. Temperatures below 10° C and above 30° C did not favour sporangial germination. *In vitro* screening of fungicides against the fungus indicated that 'Captan' 0·15% 'Thiram' 0·15% 'Perenox' 0·2% 'Ferbam' 0·2% Blue copper-50 0·3% and Ziram 0·2% completely arrested the growth of *P. nicotianae* var. *parasitica*.



PLTAE .I Fungal morphology of *Phvtophthora nicotianae* var. *parasitica* causal organism of Leaf-rot of Nagpur Oranges.

Fig. 1. Differentiation of zoospores in sporangium 1000 ×. Fig. 2. Empty sporangium after release of zoospores, 1000 ×. Fig. 3. Germinating zoospores, 450 ×. Fig. 4. Sporangiophore, 450 ×. Fig. 5. Sporangia, 500 ×. Fig. 6. Wood discolouration at infected collar. Fig. 7. Leaf-rot affected and healthy leaves of orange.

The disease condition in mandarins variously known as foot rot, collar rot or gummosis has been earlier. reported to be caused by *P. palmivora* Butler., *P. citrophthora* (Smith and Smith) Leonian in addition to *P. parasitica* Dastur [5, 7]. Leaf-fall and fruit rot reported from Mysore and Madras was caused by *P. palmivora*[4,7]. *Pythiacystis citrophthora* Smith and Smith and *P. hybernalis* Carne were found associated with a similar disease in Australia[2,8]. Attempts to isolate these species from the diseased mandarins in Vidarbha were not successful and yielded only *P. nicotianae* var. *parasitica*. This indicates that this is the only predominant species of *Phytophthora* prevalent in the gardens investigated.

Thanks are due to Dr. V.C. Lele, Mycologist, I.A.R.I., New Delhi, for kindly confirming the identity of the Nagpur isolate of *P. nicotianae* var. *parasitica*.

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---

1. Asthana, R. P., *Proc. Indian Acad. Sci.*, 1947, **24**, 243.
2. Carne, W. M., *Fruit World Australia*, 1924, **25**, 227.
3. Cole, C. F., *Jour. Agr. Victoria*, 19, p. 363.
4. Devarajan, M. B. and Aiyappa, K. M., *Ind. Farming*, 1944, **5**, 512.
5. Fraser, L., *Proc. Linn. Soc. N.S.W.*, 1949, **74**, 1.
6. Moghe, P. G., Wankar, B. N. and Kumbhare, G. B., *Nagpur Agri. Coll. Nag.*, 1970, **32**, 47.
7. Ramakrishnan, T. S., *Madras Agr. Dept. Bull.*, 1954.

BACTERIOSIS OF IRIS IN INDIA

IN Simla (1800-2100 m a. s. l.), during monsoon months, plants of iris (*Iris* sp.) were observed to develop a severe leaf blotching. The affection was particularly more prominent on rust infected leaves. Often, leaves with only a few rust pustules developed severe leaf blotch symptoms. Such leaves when viewed against light, revealed the presence of elongated, translucent, necrotic lesions in the affected parts. Microscopic examinations were suggestive of bacterial etiology of the malady.

The bacteriosis was initiated on leaves as halos of water soaked, brownish, translucent necrosis around the rust pustules. In protracted rainy weather, the infection sites expanded very rapidly and produced large, wet, elongated and brownish green patches on leaves. The lesions continued to enlarge as wet rot and covered the whole leaf surface within 5-7 days if the congenial weather conditions continued (Fig. 1). The spell of dry weather, however, arrested the development of the disease, the lesions dried up, turned dark brown and twisting of leaves occurred. In severely affected plants rhizomes were also involved and destroyed.

From young lesions, a white bacterium was isolated. The isolate produced typical symptoms of the disease when spray-inoculated on host leaves with burst rust pustules or injured artificially either by pin-pricking or celite rubbing. Infection could not occur through uninjured leaf surfaces. The bacterium was equally pathogenic to other species of *Iris* when inoculated with artificial injury. This indicated that injury to leaf surface was a prerequisite for the establishment of the infection. The ruptured leaf surface at rust pustules, thus, served as ideal sites for the initiation of the disease on iris plants, in nature. The pathogen was, however, capable of infecting gladiolus (*Gladiolus* sp.) readily without any previous artificial injuty to leaf surface.

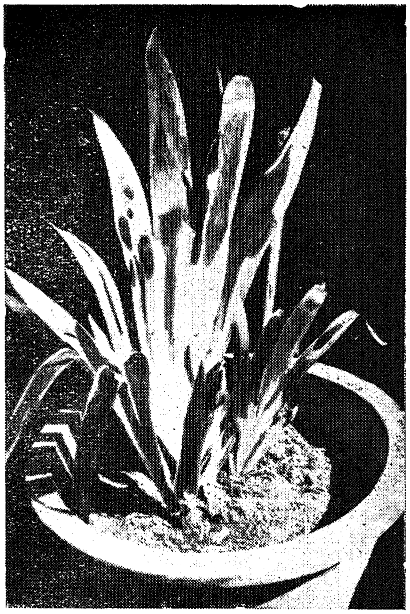

FIG. 1

The cultural, morphological and physiological characteristics of the bacterium were: white, circular, smooth, translucent, viscid, flat, glistening colonies with definite margins on NA; green fluorescent pigment produced on King's B medium; rod-shaped, Gram-negative, non-sporing, non-acid fast, motile with 1-4 polar flagella (mostly biopolar); aerobic; glucose utilized oxidatively; gelatin not liquefied; produced hydrogen sulphide (slight), indole (slight), ammonia; starch hydrolysed partially; nitrates not redueed; tolerated 5% salt in broth; asparagin utilized as a sole source of carbon and nitrogen; no peptonization and coagulation of milk; litmus reduced; produced acid without gas from galactose, glucose, mannose, glycerol but not from lactose, maltose, sucrose, salicin; utilized sodium salts of acetic, citric, lactic, propionic, tartaric but not of benzoic acids. The properties suggested that bacterium belonged to the genus *Pseudomonas*.

Of the pseudomonads reported to be parasitic to iris, *P. marginata* (McCulloch) Stapp produces a disease, broadly similar to the present one, on gladiolus and iris[1]. Our isolate also resembled *P. marginata* in most of the pathological and bacteriological characteristics[2]. The pathogen is, thus, identified as an isloate of *P. marginata*. The bacteriosis is a new record for India.

The authors wish to thank the Head, Division of Mycology & Plant Pathology, I.A.R.I., New Delhi, for providing facilities for the work.

Division of Mycology and J. C. DURGAPAL.
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New Delhi 110012, *November* 3, 1975.

---

1. Elliott, C., *Manual of Bacterial P'ant Pathogens*, 2nd ed., Chronica Botanica Co., Waltham, Mass., U.S.A., 1951. p. 52, 167.
2. Stapp, C., *Bactertal Plant Pathogens*, Oxford University Press, Amen House, London, 1961. p. 178.

NATIONAL SYMPOSIUM ON RADIATION PHYSICS

The National Symposium on Radiation Physics was held during June 10–12, 1976 at Manasagangotri, University of Mysore, Mysore. The symposium was inaugurated by Dr. R. Ramanna, Director. Bhabha Atomic Research Centre. There were in all 79 papers and 5 invited talks presented in the symposium under the following categories :

(1) Accelerators—12 papers; (2) Basic Interactions—23 papers; (3) Radiation Transport—18 papers; (4) Radiation Dosimetry—7 papers; (5) Radiation Instrumentation and Standardization—8 papers; (6) Planning of Radiation Laboratories—11 papers.

In addition there was a Panel Meeting on Smell Accelerators—their problems and prospects. In all about 12 Universities and National Laboratories participated in the symposium.

One of the highlights of this Gathering was the inauguration of the Indian Society for Radiation Physics, to provide a forum for persons working in different aspects of Radiation Physics,

SHORT SCIENTIFIC NOTES

Serum Alkaline Phosphatase Isoenzymes in Non-Laying White Leghorn Birds

Five serum alkaline phosphatase isoenzymes AKp–1 to AKp–5 including AKp–5^{S} (slow migrating) or AKp–5^{F} (fast migrating) in the order of decreasing electrophoretic mobility in laying White Leghorn birds was earlier described by the authors[1]. Savage *et al.*[2] (1970) reported that a band appeared in polyacrylamide gel electropherograms between fast and slow serum alkaline phosphatase isoenzyme in non-laying Japanese quail. No such report on serum alkaline phosphatase isoenzymes in non-laying White Leghorn is available in the literature.

The genetic variation of serum alkaline phosphatase isoenzymes was studied in fifty, clinically normal, White Leghorn pullets at five months of age by polyacrylamide disc electrophoresis[3], using *tris* boric acid EDTA buffer[4] (pH 8·8) at 150 volts, 4° C for 75 min. All gels were stained in freshly prepared staining mixture[5] at 37° C for one hour.

The pullets had a different serum alkaline phosphatase zymogram as compared to laying birds. In pullets the slow moving serum alkaline phosphatase isoenzyme (AKp–5^{S}) was absent, and a new band appeared, narrow in width, as a simple straight line which was located midway between the position of the original slow band and fast band. This new band having a greater electrophoretic mobility than the slow band (AKp–5^{S}) but less than the fast band (AKp–5^{F}) was characteristic of pullets, and has been designated as AKp–5^{P}.

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---

1. Jain, A. K., Joshi, S. C., Rawat, J. S. and Pandey, M. D., "Serum alkaline phosphatase isoenzymes and its relationship to economic traits in poultry," 1975 (Communicated).
2. Savage, T. F., Collins, W. M. and Smith, E. C., *Poult. Sci.*, 1970, **49** (6), 1622.
3. Davis, J. B., *Ann. N.Y. Acad. Sci.*, 1964, 121 (2), 404.
4. Aronsson, T. and Gronwall, A., *Scand. J. Clin. Lab. Invest.*, 1957, 9, 338.
5. Wilcox, F. H., *Genetics*, 1966, 53, 799.

Nuclear Polyhedrosis of *Plusia chalcytes* Esp. Infective to *Plusia peponis* F.

Nuclear polyhedrosis of *Plusia chalcytes* Esp. has been reported by Laudeho and Amargier (1963). Rabindra *et al.* (1975) reported the incidence of nuclear polyhedrosis in *P. chalcytes* Esp. from India. The gourd semilooper *Plusia peponis* F. has also been found to be infected with a nuclear polyhedrosis virus by Rabindra and Subramaniam (1975). The present communication deals with the nuclear polyhedrosis of *P. chalcytes* infective to *P. peponis*.

With a view to finding out the cross infectivity of nuclear polyhedrosis virus of *P. chalcytes* to *P. peponis*, 50 third instar larvae of *P. peponis* were allowed to feed on snake gourd leaves dipped in a heavy suspension of polyhedra of *P. chalcytes*. The larvae were fed with treated leaves for three days consecutively and with untreated leaves from the fourth day onwards.

It was found that the virus was infective to *P. peponis* though the mortality rate was only 40%. The incubation period was found to vary from 6–10 days. The signs and symptoms like change of colour from green to greenish white and rupturing of integument was noticed in *P. peponis* also as observed in *P. chalcytes*. The virus after a single passage through *P. peponis* was infective to the original host, *P. chalcytes* on inoculation and there was no decrease in the virulence of the virus. The virus without passage through *P. peponis* causes about 81% mortality in third instar larvae of *P. chalcytes* with an incubation period of 5–6 days.

It was noticed that the polyhedra retained the original triangular shape after passage through *P. peponis* (Fig. 1). Diameter of 50 polyhedra of *P. chalcytes* was found to vary from 0·64 μ to 1·35 μ with a mean of 0·98 μ ± 0·01 and after passing through *P. peponis* once, 50 polyhedra measured 0·66 μ to 1·68 μ in diameter with a mean of 1·05 μ ± 0·04. The retention of original shape and size of polyhedra after passage through *P. peponis* supports the positive results obtained. The inclusion bodies of nuclear polyhedrosis of *P. peponis* was found to be roughly hexagonal in shape (Fig. 1) with a diameter ranging from 0·64 μ to 2·32 μ with a mean of 1·25 μ ± 0·05. According to Gershenson (1955), the characteristic shapes and mean size of polyhedra are determined exclusively by the properties of the virus, and not by those of the host.

The authors thank Dr. Jean R. Adams, Insect Pathologist, Insect Pathology Laboratory, Beltsville, Maryland, for making the electron micrograph of the viruses.

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*1. Gershenson, S., *Mikrobiologia,* 1955, 24, 90.
2. Laudeho, Y. and Amargier, A., *Rev. Pathol. Vegetal. Entomol. Agr. France,* 1963, 42, 207.
3. Rabindra, R. J. and Subramaniam, T. R., *Curr. Sci.,* 1975, 44, 634.
4. —, Navarajan Paul, A. V., David, B. V. and Subramaniam, T. R., *Ibid.,* 1975, 44, 273.
* Original not seen.

The Scab Fungus [*Venturia inaequalis* (Cke.) Wint. on Apple Twigs in Kashmir

Recent surveys of apple orchards of Kashmir Valley to record scab incidence have revealed the occurrence of a new phase of the disease in the form of twig infection, which was hitherto unknown in India. Usually, the scab fungus infects leaves and fruits, and only in rare cases it may infect the twigs. Since the disease has assumed serious proportions in the last 2–3 years affecting almost all the apple varieties and spreading to all the apple growing areas of the three districts (Anantnag, Baramulla and Srinagar) of the Valley (Gupta and Lele, 1975), the presence of twig infection further highlights seriousness of the disease and gravity of the problem. The conidia developing in lesions on twig in spring may act as yet another source of primary infection as the ascospores produced in perithecia on overwintering leaves already known in other countries like the U.K., Israel and the European continent (Gupta, 1975).

The typical scab symptoms were observed on 1–3 years old twigs on a few apple trees of an unknown variety at Vehil (Shopian area) in June 1975. The lesions were mainly small, raised, cinnamon brown in colour with the peeling of the upper surface at the points of emergence which had silvery grey appearance. The irregular fissures covered the affected surfaces, and at places where the infection was found severe, the lesions appeared to coalesce. The infection had also resulted in the breakdown of the epidermal cells below. Microscopic examinations (the exposed surface of the lesions which were olive green to black in colour) revealed the presence of ovate to obclavate and truncate-based characteristic conidia. These were produced in indefinite numbers from each conidiophore which were annular and shades of brown. The conidiophores had arisen from the more compact stroma developed underneath the cuticle. The fungus was identified in its imperfect state as *Spilocaea pomi* Fr.

There is, therefore, an urgent need for domestic quarantine procedure to avoid the pathogen getting introduced through nursery stock from Kashmir to other hilly states, which so far, are free from this scourge of apple.

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New Delhi 110 012, *January* 14, 1976.
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1. Gupta, G. K., "Epidemiology, forecasting and possible control of apple scab [*Venturia inequalis* (Cke.) Wint.], *Pesticides,* September 1975, p. 31.
2. —, and Lele, V. C., "Fight apple scab at proper time," *Farmer and Parliament,* 11 (2 and 4), p. 17 and 21, 1976.

A New Record of *Navicula terebra* Iredale

As far as is known from the literature, only a shell of *Navicula terebra* Iredale[1] is recorded from Low Isles of Great Barrier Reef, Australia. The author came across several live specimens of this species attached to pieces of dead corals on Ajad Island (Lat. 22° 22′ 24″ N : Long. 69° 19′ 48″ E). The specimens measure on an average 8·5 mm in length, 6·0 mm in height and 3·0 mm in depth.

The present record adds a new species to the conchofauna of India. Two specimens of this species are deposited in the Museum of Marine Biological Research Station, Port Okha.

The author is grateful to Shri Moosa Raza, Fisheries Commissioner, for the facilities. His thanks are also due to Shri M. Bhaskaran for encouragement and Shri M. A. Varghese for help in identification.

Marine Biological Research Station,
Port Okha, *July* 3, 1976.
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1. Iredale, T., *Scient. Rep. Gt. Barrier Reef Exped.,* 1939, 5 (6), 293.

Sour Rot of Guava in India

A fruit rot of Guava (*Psidium guajava* L.) has been noticed in October, 1974, from orchards of College of Agriculture, Nagpur. The fungus has been identified as *Geotrichum candidum* Link ex Pers[1]. This isolate differs from the isolate of the same fungus[2], obtained from mandarin orange[3], in spore size, growth characteristics, virulence and in some physiological properties. When inoculated, the fungus produced typical symptoms of the sour rot in wounded ripe guava fruits. Mandarin orange, lemon, lime, sweet orange, banana, apple, tomato; grape-vine, watermelon, muskmelon, cucumner fruits and tubers of potato were found to develop disease symptoms on artificial inoculation with the fungus. A careful survey of literature revealed that sour rot of guava has not been yet recorded from the guava fruits on the tree.

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Nagpur, India, *July* 1, 1976.

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1. Commonwealth Mycological Institute, Collection No. IMI 190583.
2. Commonwealth Mycological Institute, Collection No. IMI 190582.
3. K. V. Shankhapal, and V. G. Hatwalne. "Sour rot of Nagpur oranges," *Indian Phytopathology*, 1974, 27 (3), 445.

Low Frequency Occurrence of the White Lac Variant in Natural Populations of *Kerria lacca* (Kerr)

The white lac variant was first recorded in a F_4 progeny from a cross of two distinct races of the common Indian lac insect *Kerria lacca* (Kerr)[1]. Two of the eight wild stocks of *K. lacca* maintained at this Institute have shown the occurrence of the white lac insect in frequencies ranging between 0·0005 and 0·001. Since deleterious recessive mutations are maintained at low frequencies in natural populations due to the combined action of mutation and selection, the white lac insect may also prove to be a recessive mutant with a selective disadvantage in its natural environment, which could reduce its utility as an insect of commerce and also as a marker gene in these insects.

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Namkum, Ranchi, Bihar, *May* 12, 1976.

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1. Chauhan, N. S. and Teotia, T. P. S., *Indian J. agric. Sci.*, 1973, 43 (12), 1086.

Occurrence of *Alternaria brassicae* (Berk.) Sacc. on *Medicago sativa* L.

A severe 'leaf-spot' disease of alfalfa (*Medicaga sativa* L.) plants was observed in September 1975, in the University Botanical Garden. The disease was noticed when the plants were six weeks old mostly on the older leaves.. Affected plants showed numerous purplish-black zonate lesions of the leaves. At the initial stages, small yellowish-brown spots are observed and soon gets enlarged. As the lesions enlarge in concentric circles, they get covered with dark sooty mould, which is easily rubbed off. Repeated isolations from these infected leaves on PDA yielded *Alternaria brassicae*. Pathogenicity studies confirmed the identification of the fungus. The culture has been deposited in the culture collection of Botany Department, Marathwada University, Aurangabad.

The fungus is known to attack mustard, cabbage, cauliflower and other Brassicaceae members[1,2]. However, there is no record of this fungus on alfalfa and this is a new host record.

Thanks are due to Prof. K. B. Deshpande of this Deparment for facilities.

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Richmond, Bangalore 560025,
June 25, 1976.

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1. Changsri, W. and Weber, *Phytopath.*, 1960, 50 (9), 631.
2. Robert McKay, *Crucifer Diseases in Ireland*, At the Sign of the Three Candles, Dublin, 1956, p. 52.

New Record of Predatory Mites on Mango Mealy Bug, *Drosicha mangiferae* Green (Margarodidae: Hemiptera)

During the survey conducted on natural enemies of insect pests of mango, two species of mites *viz.*, *Leptus* sp. and *Bochartia* sp. (Erythraeidae: Acarina) were found predating on nymphs and adults of *Drosicha mangiferae* Green in a mango Orchard at Maliahabad, Lucknow (U.P.). Later on, these predators were also recorded from other orchards in the adjoining area, highly infested with mealy bugs. Mites of both species are reddish, oval shaped, with long, six segmented, slender legs and body covered with hair like setae. Number of mites (larvae + nymphs) predating on single mealy bug ranged from 1–60. While feeding, the mites keep on tapping the body of bugs with first pair of propodosomal legs and shaking the rest with backward motion. At the point of insertion of chelicerae, the body becomes blackish while the surrounding area starts turning pale. Because of the excessive sucking of haemolymph the body gets shrivelled and the colour becomes brownish black. The mites migrate to other healthy bugs before the first prey dies and victimise them.

From January to May mites are active and all stages are found. From May onward, the adult mites migrate to soil and remain there in diapause till December. Samples of *D. mangiferae* collected from different orchards showed that 15–20% were having mites. There is no previous record of these mites as predator on *D. mangiferae* in literature, although five species of *Leptus, I.e., Leptus giganticus, Leptus poonaensis, Leptus indicus, Leptus ovatus* and *Leptus villosus* have been collected from citrus, fig and soil debris and *Bochartia* sp. has been reported predating on *Bagrada cruciferarum* KirKaldy (Khot, 1964 and Thakur *et al.*, 1969).

The authors express their thanks to the Project Coordinator (Fruits) and Head, Central Mango Research Station, Lucknow. for the facilities provided and encouragement and Director, Commonwealth Institute of Entomology, London, for identification of mites.

Central Mango Research Station,
(I.I.H.R.), B-53, Sector-A,
Mahanagar, Lucknow 6 (U.P.),
May 18, 1976.

P. L. TANDON.
BECHE LAL.

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1. Khot, N. S., *Acarologia*, 1964, 6, 681.
2. Thakur, A. V., Misra, U. S., Rawat, R. R. and Dhamdhere, S. V., *Indian J. Ent.*, 1969, 31, 86.

A Leaf Spot Disease of *Crotolaria juncea* L. Caused by *Stachybotrys atra* Corda

During the survey (September and October, 1973) of *Crotolaria juncea* in cultivated fields, a number of leaves were found infected with typical spots. The spots appeared as pale yellow turning to dark brown in colour and 5 mm in diameter. The fungus was isolated from the infected leaves on potato dextrose agar (D.D.A.) medium at 30° C. Hyphae in culture were hyaline, septate, 4–5·5 μ (dia.) with conidiophores. On the host tissue, conidiophores arise from the aerial mycelium in late September. Conidiophore bears phialides. Conidia black, smooth, elliptical, with oil drops, 8–15 μ long.

For pathogenecity test of the fungus, the spore suspension in sterile water was sprayed on healthy plants in the glass house. The inoculated plants were covered with polythene bags for 48 hours and the control maintained simultaneously. The leaves of the inoculated plants showed typical symptoms after 7 days of inoculation. Reisolations from the leaves of the artificially inoculated plants yielded the same pathogen with identical characters and identified as *Stachybotrys atra* Corda.

The fungus *S. atra* causing leaf spot disease of *Crotolaria juncea* is a first record. The culture deposited in C.M.I., England (IMI, 197813).

The authors express their grateful thanks to Dr. M. N. Gupta, Dr. K. D. Sharma, and Dr. C. P. Agarwal, for facilities and suggestions and the the Director, C.M.I. England, for confirming the identity of fungus.

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Jiwaji University, Gwalior,
May 14, 1976.

P. D. PATHAK.
R. K. S. CHAUHAN.

Balansia claviceps on Two New Hosts from Karnataka

The genus *Balansia* Speg. and its conidial stage *Ephelis* Fr. are important parasitic fungi occurring on grasses and cereals, widespread in tropics especially in India. The monographic studies on this important and interesting group of fungi have been made by Govindu and Thirumalachar (1961, 1963, 1973). In the present studies, two-additional hosts for *B. claviceps* have been reported.

Govindu and Thirumalachar (1963) reported *B. claviceps* on inflorescence of *Cyrtococcum oxyphyllum* and the present record on *C. trigonum* (Retz.) *A. camus* is a new host record. Ephelidial stage on inflorescence of *Isachne elegans* Dalz., referred to tentatively as *E. oryzae* Syd. was reported by Venkatakrishnaiya (1946). However, no *Balansia* stage has been reported on this host or related species. The present instance of *B. claviceps* on *I. dispar* Trin. is a new host record and also a new distribution. The two specimens are deposited in the Agricultural College mycological Herbarium bearing Accn. No. 1938 and 1976.

University of Agricultural Sciences,
Bangalore 560 024,
April 30, 1976.

H. C. GOVINDU.
N. SHIVANANDAPPA.

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1. Govindu, H. C. and Thirumalachar, M. J., *Mycopath. et Mycol. Appl.*, 1961, **14**, 189.
2. — and —, *Ibid.*, 1963, 20, 297.
3. — and —, *"International Syp. on Taxonomy of fungi (Abstracts), Centre for Advanced Study in Botany*, Univ. of Madras, Madras, 1973, p. 10.
4. Venkatakrishnaiya, N. S., *Curr. Sci.*, 1946, **15**, 260.

REVIEWS AND NOTICES OF BOOKS

Critical Evaluation of Systems Analysis in Ecosystems Research and Management. (Eds.). G. W. Arnold and C. T. de Wit. (Wageningen Centre for Agricultural Publishing and Documentation), 1976. Pp. 108.

The book is a collection of papers presented at the symposium on 'Critical evaluation of systems analysis and modelling in ecosystems research and management ; data collection and processing for predictive purposes', together with a few other related papers on the subject. The symposium formed part of the *First International Congress of Ecology* held during September 9–14, 1974 at the Hague, the Netherlands. There are seven papers, two of which with twin authors and the rest with single authors ; all the nine authors are directly involved in either Ecosystems or Land Resource Management research. In a sense, they are narrow specialists, though dealing with problems of widespread concern to humanity as a whole. All the same, most of the authors have touched on the meagre attention paid by the researchers on the systems analysis in ecosystems management.

All over the world people are getting increasingly concerned about the ecosystem. Studies on Man and Biosphere reciprocal effects, man-environment interactions, etc., are increasing in number and depth. During the past three decades several attempts have been made, based on statistical, and more recently, computor analyses, to predict the future of mankind on this planet and the various types of problems he is likely to encounter due to adverse effects of the changing ecosystem. While several of these predictions have not become true, the basis for such predictions have been questioned. It is this context the ecologists are resorting to find support from the advances made in physical sciences.

Application of 'systems analysis' and 'models' to biological sciences on the lines of engineering sciences, has not always resulted in accurate interpretation of the possible future. The reasons for such failures have been clearly brought out by the papers presented in this book. Some practical problems in dealing with biosphere have been listed. D. W. Goodall's illustration of the methods of subdivision of the model would benefit even a beginner in this field. The illustrative appraisal of interactions between environment, form and function by P. C. Miller and H. A. Mooney and a critical analysis of the various models suggested for grassland system by N. G. Selignan add to the value of the publication. Finally, J. N. R. Jeffer's projections for the future of systems analysis in ecology throws open several ideas, indicating that there is no finality about any model in ecology and he states 'Models are essentially transient, the product of scientific mind, and they are also the true vehicles of knowledge, so that we should talk of 'model banks' rather than 'data bank'.

In India we have just started realising the need for undertaking studies on systems analysis in ecosystems management. We should learn from the experience of the advanced countries so that we could make a more rapid progress in this field. One would get valuable information and also seed for new thoughts from this book.

G. RANGASWAMI.

Introduction to Axiomatic Quantum Field Theory. By N. N. Bogolubov, A. A. Logunov and I. T. Todorov. Authorized translation from the Russian manuscript by Stephen A. Fulling and Ludmila G. Popova, Edited by Stephen A. Fulling. (W. A. Benjamin, Inc, Reading 1975). Pp. xxvi + 707, Price: $ 32.50.

It is more than a decade since the books of Streater-Wightman and of Jost on axiomatic field theory were published. In this period, these two books have continued to be the indispensable starting point for anyone seriously interested in the more modern developments in the subject. The book under review finally provides some competition to these classics, especially in view of the fact that here again the emphasis is on the first principles of the subject rather than on its more esoteric (and, mathematically, considerably more sophisticated) recent branches. As anyone who has read the earlier book of Bogolubov and Shirkov on (a different aspect of) field theory will expect, the treatment is, within this limitation, comprehensive, thorough and exceptionally clear.

The first part is concerned with the mathematical equipment required for the rest of the book, primarily functional analysis and distribution theory. Considering that the average physicist's preparation in modern mathematics of the kind required in field theory is rather unsatisfactory, the importance of this chapter cannot be exaggerated. It is well organized and essentially self-contained though, naturally, all theorems quoted are not given proofs. Part 2 gives a detailed account of the representation theory of the Poincaré group with special emphasis on its interplay with quantum theory—this is an essential prerequisite for a proper formulation of quantum field theory. The comprehensive treatment that this part of the book is given is a welcome feature not always found in other books on this and related subjects. Part 3 gives a complete and self-contained account of the field aspects of Wightman theory while the particle aspects (scattering theory), dealt with in the next part, include the ISZ approach and Bogolubov's own approach to this question in addition to the fundamental work of Haag and Ruelle on the construction of scattering states within the framework of the Wightman theory. The results of these two parts are used, in Part, 5, to establish the well-known general theorems of field theory, including Borchers's classification of S-equivalent fields. A final part gives a very brief description of the algebraic approach of Haag and touches upon recent work in constructive field theory.

As is clear from this brief account, the book addresses itself to people who wish to have a good grounding in the foundations of modern field theory. The technique of prefacing each part with a synopsis of its contents and of covering in appropriately placed appendices specialised topics contributes greatly to the pedagogic value of the book. It is to be hoped that the authors will devote an equally distinguished successor to this volume to an equally detailed account of the more recent aspects of the subject that they have so tantalisingly skimmed over in the last part.

P. P. Divakaran.

SL_2 (IR). By Serge Lang, (Addison Wesley Publ. Co. Reading, Mass.). 1975, Pp. xvi + 428, Price $ 19.50.

It is difficult to give a satisfactory picture of the contents of this book in a few lines. The book's main object is to present the theory of infinite dimensional representations of SL_2 (IR). The first four chapters are general. In these the fundamental results in the theory of representations of compact groups, the notions of an induced representation, a spherical function, a positive definite function and their properties and their role in representation theory are presented. After this the group is SL_2 (IR). Chapters V, VI, VII, VIII centre around the proof of the Plancherel Formula. Here a self-contained account of analytic vectors, the derived representations and their classification, discrete series representations, traces of operators is given. In Chapter IX various realisations of discrete series representations and how they are related are given. Chapter X studies the universal enveloping algebra and gives a proof of the density of analytic vectors. Chapter XI constructs a special representation: the Weil representation. Chapters XII, XIII treat the decomposition of L^2 [SL_2 (Z)/SL_2 (IR)] under the action of SL_2 (IR) and Chapter XIV treats this question by a method which can be used to treat the case of a lattice instead of SL_2 (Z) [Lattice is a discrete subgroup H of SL_2 (IR) such that H/SL_2 (IR) has finite invariant volume]. This is the text. The appendice, give all the advanced result needed from functional analysis and elliptic operators: spectral theorems Sobolev's lemma, regularity theorem....

The book is rich in material and the style makes it a pleasant reading. The author has taken great pains to make it self contained. All the prerequisites are reduced to a few elementary results in Real analysis and Functional analysis. This is the best book for infinite dimensional representation theory. The first book for those who want to go further and the only book for those who want to have a good idea of the subject with proofs for SL_2 (IR). We look forward to the sequel the author promises in the preface.

V. J. Lal.

Form, Structure and Function in Plants. Edited by H. Y. Mohan Ram, J. J. Shaw and C. K. Shaw. Publishers: Sarita Prakashan, Meerut. 1975. pp. X + 457. Price: Rs. 100.

The book is a compilation of invited articles in various branches of Botany on the eve of retirement of Dr. B. M. Johri, Professor and Head of the Department of Botany in the Delhi University. The first article gives a biographical account and publications of Prof. Johri, and the remaining fifty are grouped into five sections. The papers are original articles as well as reviews from well-known authors most of whom are acquaintances of Professor Johri. Section I contains papers on Cytology and Genetics, Biochemistry; Section II on cell and tissue culture; Section III on embryology; Section IV and V on growth, morphogenesis and regeneration, and ultra structure.

It is a happy augury that on his retirement after 40 years of service in the cause of Botany, Professor Johri's colleagues and well-wishers have brought out this useful volume. The book is well-produced and contains author index and subject index, which are very necessary in a compilation of this type and will be very useful to the teachers and research workers in Botany, eventhough new information is already available on some topics.

R. Narayana.

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GROUND-STATE WAVE FUNCTION STUDIES ON Cu^{2+} ION IN TETRAKIS (PYRIDINE) COPPER (II) PEROXYDISULFATE

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ABSTRACT

A study on the ground-state wave function of Cu^{2+} ion in Tetrakis (Pyridine) Copper (II) Peroxydisulfate has been made using the available E.S.R. data. From these calculations the correct set of spin-Hamiltonian constants along with their signs and directions are picked up. These studies also gave S-contact contribution to hyperfine interaction.

THE spin-Hamiltonian constants of Cu^{2+} ion in the title complex obtained by Hecht *et al*[1], in cadmium matrix using Weill *et al*[2] method, Hecht and Frazier. III[3] in Zinc and cadmium matrices in polycrystalline form using automatic curve fitting procedure and McMillan and Smaller[5] from the second derivative of the powder and frozen solution spectra, are found to be different even though the complex studied by them was same. Since the ground-state wave function of the paramagnetic ion is very sensitive to the magnitude, sign and direction of the spin-Hamiltonian constants, it is proposed to examine the ground-state wave function for all these cases using the respective spin-Hamiltonian constants with proper signs and directions to pick up the correct set of these constants of Cu^{2+} ion in this complex. The related theory about the ground state wave function is briefly given below.

The expression for principal g and A values for rhombic crystalline field are obtained from the theory of Bleaney *et al*[6], as

$$g_x = 2 + 2/3\,\alpha(b - c) \tag{1}$$

$$\begin{aligned} A_x/P &= -K + 2/3\,(b - c) + 1/7\,(2a^2 + \gamma c - \beta b \\ &\quad - 4) \qquad (2) \\ &= -K + f_x \end{aligned}$$

Expressions, for g_y, g_z, A_y and A_z are obtained by cyclic permutations of, a, b, c and α, β, γ. With the general assumption $|xz\rangle$ and $|yz\rangle$ orbitals are degenerate one can calculate[7] the values of f_x, f_y and f_z. If proper choice is made in the assignment of directions to g_i and A_i a straight line will be obtained on the $A_i - f_i$ diagram. The slope of the line will give the value of $P(2\beta_N\beta g_N\langle r^{-3}\rangle 3d)$ and the interesection of the line of f_i axis gives the value of Fermi contact term (K). The ground-state wave functions in the case of orthorhombic symmetry for elongated case can be expressed as

$$El n = a_1\,|x^2 - y^2\rangle + a_2\,|3z^2 - r^2\rangle \tag{3}$$

where $a_1 \approx 1$ and $a_2 \approx 0$ (or $a_2 \langle\langle 1$)

Comparing these expressions with the corresponding expressions in Bleaney *et al*[6], theory, it is easy to show that $2a_2 = a + b$. It is also shown[8] that

$$a_1^2 = \frac{P_{\text{exptl}}}{P_{\text{free.ion.}}}$$

where $P_{\text{free. ion.}}$ is taken to be 360×10^{-5} cm^{-1}.

Straight lines in the $A_i - f_i$ diagram formed with the points having proper signs and directions to the spin-Hamiltonian constants are shown in Fig. 1. Different sets of spin-Hamiltonian constants obtained by three groups of workers are given in Table I with proper signs and directions along with the corresponding values $f_{//}$, f_y, f_z, P_{exptl}, K a_1^2 and a_2.

The values of P_{exptl} and K obtained from the graphs (Fig. 1 *a*) using the spin-Hamiltonian constants of Cu^{2+} ion doped in Zn and Cd complexes[3] are of the same order found in other complexes. The value of a_1 (Table I) for the above set is near unity satisfying the conditions of equation-3. For the set of spin-Hamiltonian constants obtained by Hecht *et al*[1] in single crystal form, the value of P_{exptl} (400×10^{-4} cm^{-1}) is greater than $P_{\text{free, ion.}}$ (360×10^{-4} cm^{-1}) (Fig. 1 *b*) and hence the value of a_1 is greater than unity which is obviously a violation of the conditions of equation 3. The authors themselves mentioned in their paper that there is large uncertainty in the determination of A_x and A_y values. From the set of spin-Hamiltonian constants obtained in frozen solution and powder spectra in Zn matrix[5], the values ofP_{exeti} are found to be too low (Fig. 1 *b*) and hence the values of a, is also much less than unity. From these values as a first order approximation, the spin-Hamiltonian constants obtained by Hecht and Frazier III[3] are more accurate values than the other sets. The ground-state wave functions obtained for Cu^{2+} ion using the above authors constants in Zn and Cd complexes in powder form are respectively given as

$$0{\cdot}88\,|x^2 - y^2\rangle - 0{\cdot}1225\,|3z^2 - r^2\rangle$$

$$0{\cdot}87\,|x^2 - y^2\rangle - 0{\cdot}1104\,|3z^2 - r^2\rangle$$

The S-contact contribution to the hyperfine interaction (KP) comes out to be 88 for Cu^{2+} ion both in Zn and Cd complexes.

In the above calculations we have not completely considered the exact extent of mixing of excited states, ligand orbitals and consequent perturbations on the ground-state. For taking the above aspects into consideration precise data about superhyperfine structure and optical data are required. Since in the present case unambiguous data of the above two aspects are not available we can only say that within the selected frame work of ligand theory method put forward by

TABLE I

Ground-state Parameters of Cu^{2+} ion in Tetrakis (Pyridine) Copper (II) Peroxydisulfate for different sets of spin-Hamiltonian constants

Parameter	McMillan & Smaller		Hecht & Frazier III		Hecht *et al.*
	Zn Matrix (powder form)	frozen Solution	Zn Matrix (powder form)	Cd Matrix (powder form)	Cd Matrix (single crystal)
g_z	2·169	2·204	2·252	2·264	2·264
g_y	2·056	2·061	2·059	2·071	2·0561
g_x	2·036	2·025	2·026	2·034	2·0493
$A_r \times 10^4\ cm^{-1}$	− 83	− 81	−181	−169	−184
$A_y \times 10^4\ cm^{-1}$	+ 4	− 13	− 13·5	− 13·9	− 31·5±14
$A_x \times 10^4\ cm^{-1}$	− 14	+ 11	+ 12·1	+ 15·6	56·0± 7
f_r	− 0·38	− 0·3322	− 0·2876	− 0·2749	− 0·2875
f_y	+ 0·2568	+ 0·1956	+ 0·211	+ 0·233	+ 0·3057
f_x	+ 0·3767	+ 0·4178	+ 0·407	+ 0·4044	+ 0·3364
K	0·355	0·32	0·31	0·315	0·21
$P_{expt^1} \times 10^4\ cm^{-1}$	111	124	285	280	400
$P_{eypt^1}/P_{free.\ ion}$	0·557	0·587	0·889	0·878	1·05
a_2	− 0·0738	− 0·1366	− 0·1225	− 0·1104	− 0·0200

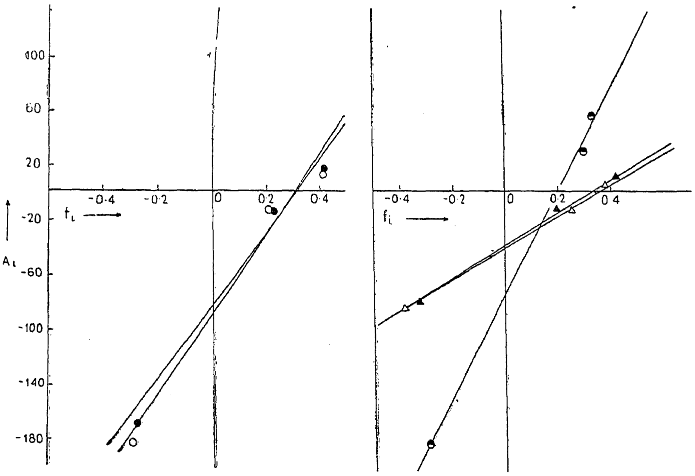

FIG. 1. $A_i - f_i$ diagrams of Cu^{2+} ion in Tetrakis (Pyridine) Copper (II) Peroxydisulfate. (*a*) Hecht & Frazier III ● ● ● Cd Matrix, ○ ○ ○ Zn Matrix. (*b*) (1) McMillian & Smaller, ▲ Frozen Solution △ Zn Matrix. (2) Hecht *et al.*, Cd Matrix.

Bleaney *et al.*, the spin-Hamiltonian constants obtained by Hecht and Frazier III[3] are accurate values.

---

1. Hecht, H. G., Kerns, R. C. and Frazier III, J. P., *J. Inorg. Nucl. Chem.*, 1969, **31**, 1983.
2. Weill, J. A., Goodman, G. L. and Hecht, H. G., *Paramagnetic Resonance*, Edited by W. Low, Pp. 880–895, Academic Press, New York, 1963.
3. Hecht, H. G. and Frazier III, J. P., *J. Chem. Phys.*, 1966, **44**, 1718.
4. Johnston, T. S. and Hecht, H. G., *J. Mol. Spectry.*, 1965, **17**, 98.
5. McMillan, J. A. and Smaller, B., *J. Chem. Phys.*, 1961, **35**, 763.
6. Bleaney, B., Bowers, K. D. and Pryce, M. H. L., *Proc. Roy. Soc.*, 1955, **228**, 166.
7. Kokoszka, G. F., Allen, H. C. and Gorden, G., *J. Chem Phys.*, 1967, **42**, 3020.
8. Chadda, C., *J. Phys. Soc.*, Japan, 1968, **24**, 976.

SEQUENCE DISTRIBUTION IN ACRYLONITRILE-METHYLMETHACRYLATE RANDOM COPOLYMERS BY N.M.R. SPECTROSCOPY

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ABSTRACT

The sequence distribution studies on the acrylonitrile-methylmethacrylate copolymer of high methylmethacrylate (M) content ($30\% < M < 75\%$) by NMR studies is reported in this paper. A tentative assignment has been made for the isolated M centered unit AMA, 2M centered units MMA (or AMM) based on the shape of the peaks observed for methoxy and α-methyl protons. The presence of MMM type of triad sequences is indicated by the pattern of α-methyl protons.

INTRODUCTION

NMR is one of the most important techniques for the determination of sequence distribution in polymer and coplymer systems. Using this method, the syndiotactic, isotactic and heterotactic tried distribution in polymethylmethacrylate (PM) and polyacrylonitrile (PA) have been studied in detail by a number of workers[1-9]. However, not much work has been done in the case of acrylonitrile-methylmethacrylate (A–M), copolymers. The work of Pham[10] in this system is mainly confined to copolymer samples of low methylmethacrylate (M) content ($1\% \leqslant M \leqslant 20\%$)· Hence, an attempt has been made to determine the sequence distribution of A–M copolymers of high M content and also to determine the % composition of these samples and to compare with the values obtained by the chemical analysis.

MATERIALS AND METHODS

A–M copolymers of four different compositions (MA_1, MA_2, MA_3 and MA_4) were prepared by free radical polymerisation using benzoyl peroxide (0·05 mole %) as initiator[11] and the percentage of conversion was about 20. Table I gives the monomer feed, the percentage of conversion and the mole% of A· A content was analysed by N_2 analysis.

The NMR spectra were recorded by a varian HA-100 Spectrometer at 120 °C. Deuterated dimethylsulfoxide (*d*6–DMSO) was used as solvent. The concentration of the solutions usedis 10%. Tetramethylsilane (TMS) is used as internal reference. Figure 1 shows the spectra of copolymers, as well as of the parenthomopolymers.

RESULTS AND DISCUSSION

The spectrum of pure PM (Fig. 1) has the following peaks. The methoxy protons of PM homopolymers[2,10,3] in *d*6–DMSO resonate at 6·27 τ. The methylene protons appear at 8·05 τ and 8·11 τ respectively; α-methyl protons appear between 8·6–9·4 τ, which consists of three peaks at 8·78 τ, 8·95 τ and 9·11 τ corresponding to isotactic, heterotactic and syndiotactic triad sequences respectively.

The spectrum of PA homopolymer shows a quintuplet from methine (CH) protons centered at 6·72 τ. Methylene protons appear as a complex pattern in the range 7·4–8·0 τ. This has been attributed to the combination of two isotactic and syndiotactic diads[1,4,10] centered at 7·76 τ and 7·79 τ respectively.

In the A–M copolymer systems (Fig. 1) four groups of peaks are observed, as noticed by Pham[10]. The peaks in the range 5·8–6·4 τ corresponding to

TABLE I

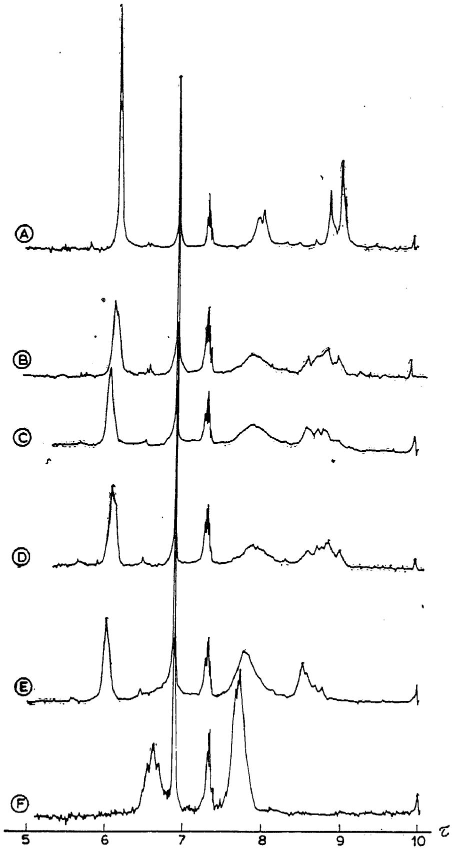
Initiator: Benzoyl peroxide

Solvent: DMF.
Temperature: 60 ± 0·05° C

Copolymers	Mole % of A in Monomerfeed	% of Conversion	Composition[a]		Composition[b]	
			A%	M%	A%	M%
MA_1	37·6	20·2	28·9	71·1	28·9	71·1
MA_2	57·2	10·4	41·5	58·5	42·2	57·8
MA_3	74·6	21·9	56·6	43·4	54·1	45·9
MA_4	84·2	17·6	65·7	34·3	65·1	34·9

[a] by N_2 analysis, [b] from NMR.

methoxy protons of the copolymers appear at a slightly lower field as compared to that of PM. It is also noticed that as the composition of M is increased, the peaks move to slightly higher field.



NMR SPECTRA AT 100 MHz. Ⓐ-PMMA, Ⓑ-MA1, Ⓒ-MA2 Ⓓ-MA3, Ⓔ-MA4 AND Ⓕ-PAN

FIG. 1

The quintuplet observed in the case of PA homopolymer for methine (CH) protons is not present in all the polymer samples studied ($M \leqslant 20\%$). In fact, these peaks diminished when the mole% of M was increased from 4·4 to 20·0 in the systems studied by Pham[10].

The peaks between 7·6 and 8·5 τ are due to methylene protons of the copolymers. All these peaks appear as a broad band. The peaks move from lower to higher field as M content is increased.

The α-methyl protons resonate in the range 8·4 to 9·2 τ. These peaks appear at a slightly lower field as compared to peaks of α-methyl protons of PM.

The shift noticed corresponding to methoxy and α-methyl protons for the spectra recorded in d7-DMF is different from what is observed for the copolymers in *d*6-DMSO. This may be due to the solvent effect. Absorptions near 7·46 τ, and those in the range of 7·34–7·08 τ are due to hydrogen atoms remaining in *d*6-DMSO and to water absorbed in the solvent respectively[1].

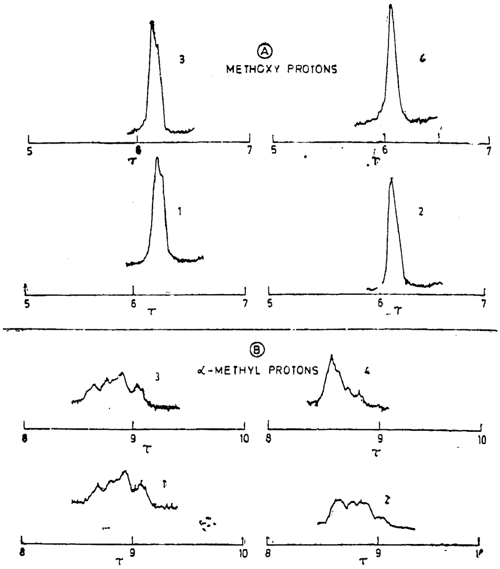


FIG. 2 NMR SPECTRA AT 100 MHz (1) MA1, (2) MA2, (3) MA3 & (4) MA4

The α-methyl proton peaks of the coplymers (Figs. 1 and 2) are a complex combination of different types of triad sequences, as observed by Pham[10]. The peak in all the copolymer systems (in *d*6-DMSO) appears as a broad one and this broadening increases as the M content is increased in the copolymer as pointed out by Pham. These α-methyl peaks comprise of triad sequences of isolated M centered units (*i.e.* AMA type). Pham[10] observed a shoulder at 8·83 τ (*i.e.*, 1·17 δ value) which he related to 2M centered MMA (or AMM) triad sequences. He expected that the intensity of this peak might increase as the composition of M increases in this system. If this is the case, shoulder appearing at 8·71 τ value in MA$_4$ might be the same type of 2M centered sequences, but for the other copolymers, it is difficult to say anything about these sequences from the spectrum. Since the composition of M in all the copolymers are high, it is prossible to assume that the 2M centered MMA (or AMM) type sequences appear in abundance in addition to the triad sequences of MMM type. The reason for this assumption is that the broad spectrum of α-methyl peaks is slowly shifted to higher field as M content is increased, where the α-methyl protons of PM appear. In addition to that a gradual increase of the peaks connected to syndiotactic and heterotactic type of peaks observed in PM is also noticed.

The methoxy protons peaks of the copolymer (Figs. 1 and 2) correspond to traid distribution of the type

AMA, whereas a pronounced shoulder in all the methoxy peaks represents the configurational triad distribution of 2M centered MMA (or AMM) type of units. This shoulder becomes less pronounced as M content is decreased. Triad sequences of the type MMM are not observed clearly in these peaks, which should appear at a slightly higher field (*i.e.*, at $6 \cdot 27\ \tau$). These observations are in conformity with those made by Pham[10]. But, from the α-methyl proton peaks, it was assumed that triad sequences of MMM type are present. This can be clarified by obtaining spectra with better resolution or by simulting these peaks. This work is in progress.

The composition of the copolymers by NMR analysis has been determined by the method of Pham[10] using the following relation

$$\text{Mma\%} = \frac{(1/3)\ \text{Integral of Methoxy (or } \alpha\text{-methyl) protons}}{(1/2)\ \text{Integral of methine protons.}}$$

These values are given in Table I. They agree well with the values obtained by N_2 analysis.

ACKNOWLEDGEMENT

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1. Matsuzaki, K., Uryu, T., Ishigure, K. and Yakeunchi, M., *Polymer Letters*, 1966, **4**, 93.
2. Bovey, F. A., *NMR Spectroscopy*, Academic Press, New York and London, 1969, p. 168.
3. Preprints of lectures in *Spectroscopy of Polymers*, Prague, 1967, Summer School, VI/11.
4. Beevers, R. B., *Macromolecular Reviews*, 1968, **3**, 202.
5. Bovey, F. A. and Tiers, G. V. D., *J. Polymer. Sci.*, 1960, **44**, 173.
6. Coleman, B. D., Fox, T. G. and Reinmoller, M., *Ibid.*, Pt. B, 1966, **4**, 1029.
7. Matsuzaki, K., Uryu, T., Ishida, A. and Ohki, T., *Ibid.*, Pt. B, 1964, **2**, 1139.
8. Yamadere, Rand Murano, M., *Ibid.*, Pt. B, 1965, **3**, 821
9. Matsuzaki, K., Uryu, T., Okada, M., Ishigure, K., Ohki, T. and Yakeunchi, M., *Ibid.*, Pt. B, 1966, **4**, 487.
10. Pham, Q. T., *NMR—Basic Prin ciples and Progress*, Springer-Verlag, Berlin-Heidelberg, New York, 1971, Vol. 4, pp. 119–128.
11. Kashyap, A. K., Reddy, C. R. and Kalpagam, V. *J. Ind. Chem. Soc.*, 1976, **53**, 106.

THE DEFORMATION BEHAVIOUR OF S.G. IRONS

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ABSTRACT

The deformation behaviour under compression at room temperature of cast and ferritised S.G. (Spheroidal Graphite) irons has been studied to understand their high ductility. It has been established that on deformation the spheroidal graphite particles distort considerably and take to ellipsoidal shapes. The deformability index values suggest that graphite, although brittle by itself, deforms to a greater extent than either ferrite or pearlite in the matrix. It has also been found that an increase in strain decreases the deformability index of graphite.

S. G. IRONS also known as nodular irons or ductile irons, are extensively used in industry[1,2]. They are stronger than gray cast irons and their superior strength is understood in terms of the smaller graphite (crack) length. Their good ductility is, however, not well understood.

It is now known that the ductility of two-phase materials is mostly dependent on the volume fraction and shape and is independent of the nature, strength or size (in the range of 1 to 200 μ) of the second phase particles[3,4]. The mechanism of fracture is, however, different for two-phase materials containing hard and soft particles. The former gives rise to stress concentrations resulting in the formation of holes in their vicinity. If the second phase, particles are soft, they undergo equal or more deformations than the specimen as a whole.

The amount of deformation undergone by the dispersed phase in comparison with that of the specimen is expressed in terms of the deformability index, ν, given by

$$\nu = \frac{\text{amount of deformation undergone by the second phase}}{\text{amount of the deformation undergone by the specimen as a whole}}$$

Malkiewioz and Rudnik[5] showed that the deformability index for tensile stress conditions is given by

$$\nu = \frac{2 \log (b/a)}{3 \log (l_f/l_0)}$$

where l_0 and l_f are the initial and final lengths of the specimen and b and a are the major and minor axes of the ellipsoidal shape of the inclusion obtained after deformation.

It can easily be shown that for conditions of compression the equation is modified to

$$\nu = \frac{2 \log (b/a)}{3 \log (l_0/l_f)}$$

It has been found that most of the inclusions in steel have got ν values of $0 \leqslant \nu \leqslant 1$ at the high temperatures of working[6]. The deformation behaviour of S.G. Irons in the as-cast and ferritised conditions has been studied in the present investigation with a view to obtain information on their ductility characteristics.

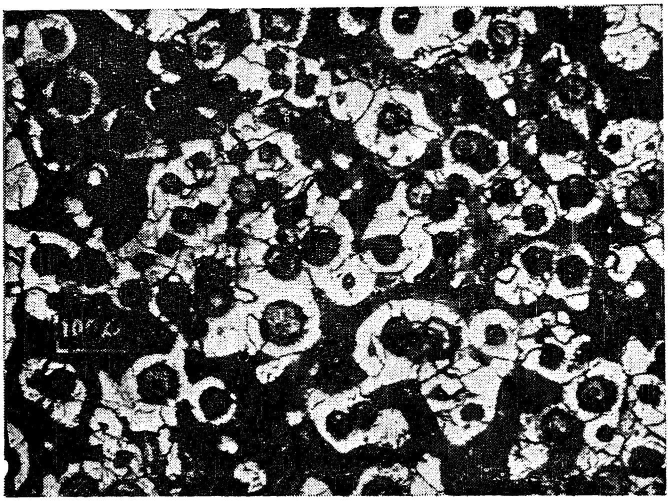

FIG. 1. Optical micrograph of the as-cast S.G. Iron.

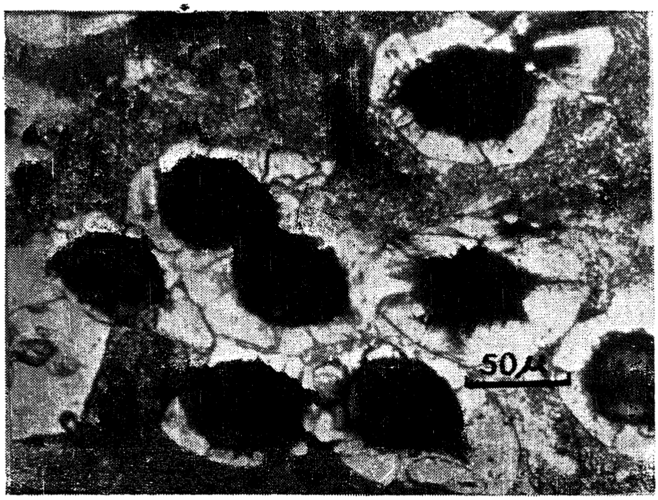

FIG. 2. Optical micrograph of the cast S.G. Iron after 25% deformation.

EXPERIMENTAL PROCEDURES

S.G. Iron of composition 3·4% carbon, 1·9% silicon, 0·46% manganese was used in the present work. Specimens of 6 × 6 × 10 mm were studied in the as-cast condition and also after giving a ferritising treatment. For the latter, specimens sealed in a silica tube were heated at 920° C for 1 hr, slowly cooled in the furnace to 700° C and were held at this temperature for 11 hours before cooling slowly to room temperature. The specimens were prepolished and etched on one of the longer surfaces and compressed perpendicular to the polished surface to various extents (6 to 25%) in an Amsler 30 ton Universal Testing Machine. The deformed specimens were examined in an optical microscope for noting the changes in microstructure.

RESULTS AND DISCUSSION

Figure 1 is the optical microstructure of the as-cast S.G. iron showing the graphite nodules encapsulated in ferrite zones lying within a pearlite matrix. The micrograph in Fig. 2 of the specimen compressed to

25% reduction reveals that the spheroidal graphite has become ellipsoidal on deformation. Figure 3 is the optical micrograph of the ferrite matrix. On deforming 23% the ferritic S.G. iron ,shows the distorted graphite modules as well as the considerable deformation undergone by the matrix Fig. 4.

The deformability index values (ν) calculated from quation (2) are shown in Table I. The ν values for all deformations studied in both ferritic and as-cast states are greater than unity and this value decreases steadily with increasing strain. This suggests that graphite deforms more readily than the matrix on the initial stages of deformation.

It is interesting to note here that the graphite nodules undergo severe distortion when S.G. irons are deformed. This is not normally expected as graphite has a layer-type hexagonal structure and a low surface energy of bonding to the matrix, and polycrystalline graphite is soft but brittle[7]. It may be assumed that the very low tensile strength of graphite coupled with the low bonding energy makes graphite to act like voids. Holes in materials can also be considered as second phase particles and a spherical hole tends to elongate and flatten into an ellipsoidal shape on deformation[6]. It is then difficult to understand the variation of the deformability index with the degree of compression,

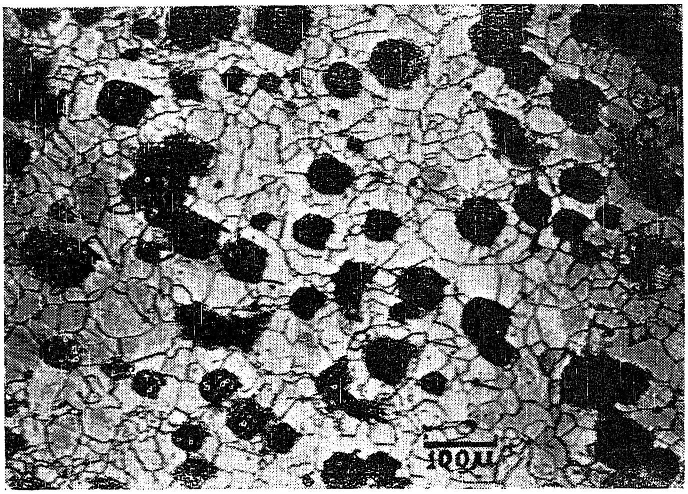

FIG. 3. Optical micrograph of the ferritised S.G. Iron.

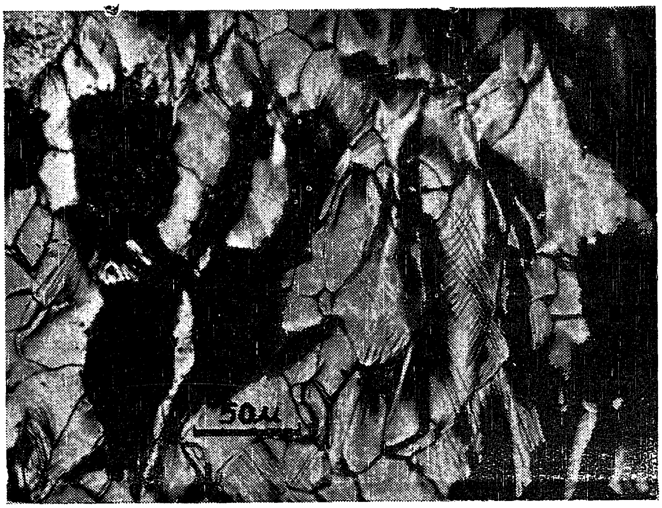

FIG. 4. Optical micrograph of the ferritised iron after 23% deformation.

TABLE I

Deformability Index Values of Graphite in the As-Cast and Ferritised S.G. Irons

Treatment	% deformation	Deformability index
As-cast	6	2·8
As-cast	13	2·0
As-cast	25	1·0
Ferritised	8	2·3
Ferritised	15	1·5
Ferritised	23	1·0

The ductile behaviour of the S.G. iron can therefore be attributed to the ease with which the graphite nodules change shape on deformation. This is not possible in the gray cast irons, since the graphite flakes act as stress raisers and do not deform with the metal.

CONCLUSION

The graphite nodules of S.G. iron deform along with the matrix and the nodular shape changes to ellipsoidal shape on deformation. The calculated deformability index values show that the relative magnitude of deformation of graphite with respect to the matrix changes with the degree of strain.

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1. Heine, R. W., Loper, C. R. and Rosenthal, P. C., *Principles of Metal Casting*, McGraw-Hill (New York), 1967, p. 614.
2. Lyman, T. (ed.), *Metals Handbook*, American Society for Metels (Ohio), 1961, p. 379.
3. Mogfield, I. L., *Metallurgical Reviews* (incorporated in Metals and Materials), 1967, **12**, 49.
4. Rosenfield, A. R., *Ibid.*, 1968, **13**, 29.
5. Malkiewicz, T. and Rudnik, S., *J. Iron and Steel Inst.*, 1963, **201**, 33.
6. Kiessling, R., *Non-metallic Inclusions in Steel*, Iron and Steel Inst. Publication (London) No. 115, 1968, p. 54.
7. Decker, R. F., *Metallurgical Trans.*, 1973, **4**, 2495.

ROLE OF MAGNESIUM AND IRON IN THE REVERSAL OF NICKEL TOXICITY IN *OGAWA* SEROTYPES OF *VIBRIO CHOLERAE* AND *VIBRIO ELTOR*

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ABSTRACT

The effect of different concentrations of Ni^{2+} on growth, acid production and glucose utilisation of *Ogawa* serotypes of *Vibrio cholerae* and *Vibrio eltor* has been studied. Ni^{2+} was toxic to both, particularly for acid production. The reversibility of Ni^{2+} toxicity by Fe^{3+} and Mg^{2+} showed that Ni^{2+} induces a conditioned Mg^{2+} deficiency in both *V. cholerae* and *V. eltor*.

INTRODUCTION

THOUGH nickel has been placed with "unfamiliar trace elements" from the point of view of microbial nutrition[1], evidence as to its essential function is gradually accumulating[2]. For instance, Abelson and Aldous[3] demonstrated that excess of nickel and other heavy metals interfere with magnesium metabolism in microorganisms and Healy *et al.*[4] observed that nickel reduced succinic dehydrogenase but doubled catalase activity in *Neurospora crassa*. Interestingly, whereas magnesium was effective in counteracting nickel toxicity in *Aspergillus niger*[5] and *Neurospora crassa*[6], zinc was effective in *Chlorella vulgaris*[7] and both iron and magnesium proved effective in reversing nickel toxicity in *Micrococcus pyogenes* var. *aureus*[8].

In *Inaba* serotypes of *Vibrio cholerae and Vibrio eltor*, Karuna Sagar *et al*[9] noted that whereas iron was more effective in counteracting nickel toxicity in the former, magnesium was in the latter. In this communication we present the results of our experiments with *Ogawa* serotypes of *Vibrio cholerae* an *Vibrio eltor*.

MATERIALS AND METHODS

One strain of *Vibrio cholerae Ogawa* (NCTC 8021) and one strain of *Vibrio eltor Ogawa* (NCTC 10255) obtained from Central Public Health Laboratory, London, were used.

The test medium of pH 7·6 contained tryptone (Oxoid) 1%, sodium chloride (BDH) 0·5% and glucose (BDH) 1%. Solutions of nickel sulphate (May and Baker Ltd.), ferric ammonium citrate (Merck) and magnesium sulphate (Merck) were prepared to contain 1000 mcg of metal per ml.

Ni^2 solution was added aseptically to the tubes containing 10 ml test medium to provide the metal concentrations ranging from 50 to 1000 mcg per tube Experiments conducted to determine toxicity reversals contained Ni^{2+} sufficient to induce 50% growth inhibition and reversing metals, Fe^{3+} / Mg^{2+} in concentrations from 50 to 1000 mcg per tube. Aliquots of 0·05 ml of 20 h peptone water culture of vibrios containing approximately 10^8 viable cells were used to inoculate the tubes. After incubation for 24 h, growth was measured colorimetrically at 660 nm. Glucose utilisation was determined by following its disappearance from the medium by the technique of Folin and Wu[10]. Acid production was estimated by titrating 2 ml aliquots of culture medium against 0·005 N NaOH using bromothymol blue as indicator.

All the experiments were repeated four times and the results were consistent. Average values were expressed as percentage of values recorded for control culture.

RESULTS

Table I shows the effect of Ni^{2+} on *Ogawa* serotypes of *Vibrio cholerae* and *Vibrio eltor*. Complete inhibition of growth occurred at 600 mcg in the former and 500 mcg in the latter. However, 50% growth inhibition was noted at 200 mcg level in both the biotypes. The fall in acid production was more in *V. cholerae* than in *V. eltor*. Thus at 200 mcg which produced 50% growth inhibition in both biotypes, acid production was seen to the extent of 25% in *V. cholerae* and 44% in *V. eltor*.

Table II shows the effect of Fe^{3+} and Mg^{2+} supplementation to *V. cholerae* culture containing a concentration

TABLE I

Effect of Ni^{2+} *on* Ogawa *serotypes of* V. cholerae *and* V. eltor (*Values expressed as percentage of control*)

	Concentration of Ni^{2+} (mcg/10 ml medium)									
	Nil	50	100	150	200	250	300	400	500	600
					V. cholerae					
Growth	100	83	65	58	50	44	44	44	39	..
Acid production	100	50	50	30	25	12	12	12	12	..
Glucose utilisation	100	100	80	70	70	50	50	50	50	..
					V. elter					
Growth	100	90	70	55	50	35	25	20	..	..
Acid production	100	62	60	44	44	20	12	12	..	..
Glucose utilisation	100	80	80	80	60	60	40	40	..	..

TABLE II

Effect of Mg^{2+} *and* Fe^{3+} *in presence of* 200 *mcg of* Ni^{2+} (50% *growth inhibition*) *in* V. cholerae Ogawa (*values expressed as percentage of control*)

	Control	Nil	Concentration of Mg^{2+} / Fe^{3+} (mcg/10 ml medium)												
			50	100	150	200	250	300	400	500	600	700	800	900	1000
						Ni (200 mcg) + Mg									
Growth	100	50	58	70	80	80	80	80	80	80	80	89	95	95	95
Acid production	100	25	40	45	50	50	55	55	65	75	75	75	75	75	75
Glucose utilisation	100	70	70	70	80	100	100	100	100	100	100	100	100	100	100
						Ni (200 mcg) + Fe									
Growth	100	50	50	50	50	50	50	50	50	50	50	50	50	50	50
Acid production	100	25	25	25	5	25	25	25	25	25	25	25	25	25	25
Glucose utilisation	100	70	70	70	70	70	70	70	70	70	70	70	70	70	70

TABLE III

Effect of Mg^{2+} and Fe^{3+} supplementation on Ni^{2+} inducing 50% *growth inhibition in* V. eltor Ogawa (*values expressed as percentage of control*)

	Control	Nil	50	100	150	200	250	300	400	500	600	700	800	900	1000
			Concentration of Mg^{2+} / Fe^{3+} (mcg/10 ml medium)												
						Ni (200 mcg) + Mg									
Growth	100	50	68	68	68	70	78	78	84	84	84	84	84	84	84
Acid production	100	44	50	50	50	50	50	50	50	60	60	60	60	60	60
Glucose utilisation	100	60	60	60	60	80	100	100	100	100	100	100	100	100	100
						Ni (200 mcg) + Fe									
Growth	100	50	50	50	50	50	50	50	50	50	50	50	50	50	50
Acid production	100	44	40	40	32	22	22	22	22	18	18	18	18	18	18
Glucose utilisation	100	60	60	60	60	55	55	50	50	50	50	50	50	50	50

of Ni^{2+} (200 mcg) inducing half maximal growth. Mg^{2+} was partially effective in reversing Ni^{2+} toxicity. Growth reached a maximum value of 95% at 800 mcg level. Though normal utilisation of glucose was observable at 200 mcg, no increase in acid production occurred beyond 75%. Fe^{3+} was completely ineffective in reversing Ni^{2+} toxicity.

A similar situation was evidenced in the case of *V. eltor* also (Table III). Supplementation of Mg^{2+} brought about a maximum of 84% in growth at 400 mcg. Glucose utilisation reached the control value at 250 mcg but optimal acid production was only 60% (500 mcg). Fe^{3+} was not only ineffective in reversing Ni^{2+} toxicity, but was in fact inhibitory, particularly to acid production.

DISCUSSION

The above results clearly show that Ni^{2+} was toxic to both the biotypes of vibrios. Though half maximal growth was attained at the same concentration of Ni^{2+} in both the biotypes, a greater inhibition of acid production was noted in *V. cholerae.*

Mg^{2+} was partially effective in reversing the toxicity due to Ni^{2+} in both the biotypes. Though glucose utilisation was restored in both *V. cholerae* and *V. eltor* acid production was still low. Abelson and Aldous[3] indicated that the primary influence of Mg^{2+} is a control on the toxic metal uptake which presumably results in the depression of intracellular concentration of toxic ions to non-toxic levels. If their surmise is right it would be natural to expect all adverse influence of metal toxicities to be simultaneously annulled. Since this does not happen in Ni^{2+} toxicity control of ion uptake is not perhaps the only mechanism of reversal of metal toxicity by Mg^{2+}. In fact, Adiga *et al*[5] and Narasimha Rao and Nagesha[8] arrived at similar conclusions from their studies on toxicity reversals.

Fe^{3+} totally failed to alleviate Ni^{2+} toxicity. This is interesting from the viewpoint of the observations of Narasimha Rao and Nagesha[2] and that of Adiga *et al*[5] that both Fe^{3+} and Mg^{2+} were effective in reversing Ni^{2+} toxicity. Our results indicate that Ni^{2+} might be inducing a conditioned deficiency of Mg^{2+} in *Ogawa* serotypes of *V. cholerae* and *V. eltor.*

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1. Hutner, S. H., *Ann. Rev. Microbiol.*, 1972, 26, 313.
2. Bartha, R. and Ordal, R. J., *J. Bacteriol.*, 1965, 89, 1015.
3. Abelson, P. H. and Aldous, E., *Ibid.*, 1950, **60**, 401.
4. Healy, W. B., Cheng, S. and Mc Elroy, W. D., *Arch. Biochem. Biophys.*, 1955, **54**, 206.
5. Adiga, P. R., Sastry, K. S., Vankatasubramaniam, V. and Sarma, P. S., *Biochem. J.*, 1961, 81, 545.
6. Sivarama Sastry, K., Adiga, P. R., Venkatasubramaniam, V. and Sarma, P. S., *Ibid.*, 1962, 85, 486.
7. Upitis, V. V., Pakolne, D. S. and Nollendorf, A. F., *Mikrobiologia*, 1973, **42** 758.
8. Narasimha Rao, S. and Nagesha, C. N., *Indian J. Expt. Biol.*, 1967, **5**, 182.
9. Karuna Sagar, I., Narasimha Rao, S. and Nagesha, C. N., *Ibid.*, 1976, **14**, 51.
10. Folin, O. and Wu, H., *J. Biol. Chem.*, 1920, **41**, 367,

PLAQUE FORMATION OF ARBOVIRUSES UNDER AN OVERLAY OF TISSUE CULTURE MEDIUM AND LIQUID PARAFFIN

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THE plaque technique provides a sensitive and accurate method for the quantitative estimation of virus infectivity in cell cultures. This technique was first introduced by Dulbecco in 1952 for animal viruses. Since then it is widely employed in the field of virology with several modifications though the principle of the original technique essentially remains the same. Most viruses require solid overlay medium to produce plaques and agar is commonly used in such studies. However, substances such as agarose, methylcellulose[2], carboxymethyl cellulose[3], startch[1], Tragacanth gum[4] or any other substance which inhibits the production of secondary plaques can also be used for the formation of plaques. A viscous substance is usually added to overlay medium which solidifies or remains viscous. Vaccinia is the only virus which produces clear plaques even under liquid tissue culture medium[5,6]. In the present study formation of plaques of two arboviruses under an overlay consisting of a combination of liquid tissue culture medium and liquid paraffin was successfully carried out.

Vero cell culture was employed and the tests were conducted in linbroplates. Vero cell monolayers were grown in wells of linbroplates employing MEM (Glasgow) with Earle's base and 5 per cent goat serum (G.S.).

Commercially available liquid paraffin (LPFN) obtained from Agenta Chemicals, Bombay, India and MEM double strength with 10% G.S. were used in the overlay. 0·1 per cent Amido black[3] was employed for staining the cells. Tissue culture stocks of two arboviruses: Sindbis (Group A) and Chandipura (Vesicular Stomatitis group) were studied.

The method followed for conducting the experiments was as follows:

(*a*) The medium was removed from the wells of linbroplate with confluent monolayers of Vero cell culture and dilutions of virus were inoculated in 0·1 ml quantities to each well and adsorbed for one hour at 35° C with intermittent rocking of the plates.

(*b*) After adsorption MEM double strength with 10% G.S. was added in 0·5 ml quantities followed by one ml quantities of LPFN into each well. MEM double strength was used instead of MEM as the cell sheet was degenerating faster when the latter medium was employed. The LPFN formed a separate layer on top of the liquid tissue culture medium. While growing Vero cell monolayers and after virus inoculation, the plates were incubated at 35° C in carbon dioxide incubator. Results were noted by staining with 0·1 per cent Amido black on appropriate post-inoculation days as determined by earlier experiments. Sufficient precaution was taken to avoid unnecessary handling and movement of the test plates during incubation to avoid development of secondary plaques.

The size and titre of the plaques of the two viruses (Sindbis and Chandipura) are presented in Table 1. Figures 1 and 2 show plaques of these two viruses.

TABLE I

Virus	Overlay	Day of appearance of plaque	Plaque titre log$_{10}$, PFU/ML.	Plaque diameter
Sindbis (AR 339 Gr. A,	Liquid paraffin	3	7·3	Micro-plaques
Chandipura (653514) (Vesicular stomatitis group)	do.	1	8·3	2 mm

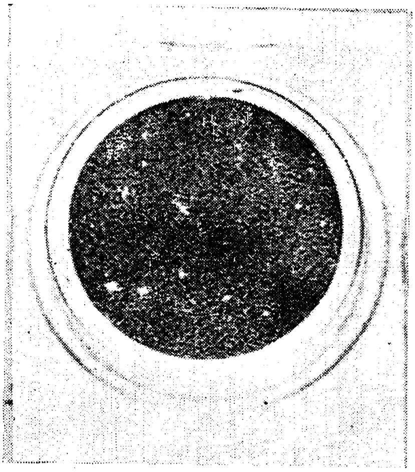

FIG. 1. Plaques of Sindbis Virus.

Plaques were produced in our experiments by employing a combination of liquid tissue culture medium and LPFN in overlay. However, when overlaying was done with only liquid tissue culture medium, cytopathogenic effect was produced. LPFN alone in the absence of liquid tissue culture medium failed to produce plaques. The mechanism responsible for the formation of plaques under this overlay is the pressure exerted by the layer of liquid paraffin which prevents the rapid spread of infective foci that results in the formation of plaques.

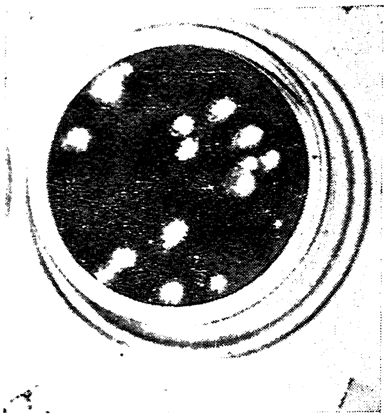

FIG. 2. Plaques of Chandipura Virus.

The advantage of plaquing under this overlay is that it permits the virus to form plaques in liquid tissue culture medium itself and the effect of LPFN is only indirect on the plaques. The easy availability of LPFN makes it an useful adjunct to plaque study.

ACKNOWLEDGEMENT

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1. De Maeyer, E. and Schonne, E., *Virology*, 1964, **24** 13.
2. Hotchin, J. E., *Nature*, 1955, **175**, 352.
3. Madrid, A. T. and Porterfield, J. S.. *Bull. Wld. Hlth. Org.*, 1969, **40**, 113.
4. Mirchamsy, H. and Rapp, F., *Proc. Soc. Exp. Biol. Med.*, 1968, **129**, 13.
5. Postlethwaite, R., *Virology*, 1960, **10**, 466.
6. Slonim, D. and Hulenova, M., *Acta Virol.*, 1969, **13**, 329.

PHYSIOLOGICAL STUDIES ON THE EFFECTS OF NUTRITIONAL IMBALANCE ON THE CENTRAL NERVOUS SYSTEM: EFFECTS OF THIAMINE DEFICIENCY ON THE REGIONAL PROTEIN METABOLISM IN THE BRAIN OF CHICKEN, *GALLUS DOMESTICUS*

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ABSTRACT

The levels of RNA, protein and the activity levels of Aspartate amino-transferase (AAT) and Alanine aminotransferase (AlAT) in the cerebrum, cerebellum and medulla oblongata of thiamine-deficient chicken increased indicating an augmentation in the activity of the protein synthetic machinery in B_1-hypovitaminous birds.

THE proteins in the brain of vertebrates are known to exist in a dynamic state[1]. The synthesis and catabolism of proteins in the mammalian brain have been extensively studied[2-4]. It has been demonstrated that the levels of protein, total nitrogen and DNA remain practically unchanged, even in states of severe protein deficiency or starvation[6-8].

However, information about the regional distribution and metabolism of protein in the vertebrate brain during thiamine deficiency is lacking. Since marked changes in the enzymes connected to carbohydrate metabolism occur in animals with thiamine deficiency[5,9], it might be expected that the brain would also show alterations connected to protein metabolism during thiamine deficiency. Little work has been done to test this possibility. Hence, the present study was undertaken.

MATERIAL AND METHODS

Two days old, male white leghorn chicken, *Gallus domesticus* (10–12 g) were reared in the laboratory at 36 ± 2° C. The controls were fed on standard chicken feed. The experimental animals were fed on polished rice to induce thiamine deficiency[5]. Water was available *ad libitum* to both the groups.

The normal and experimental birds were decapitated after 28 days. The brain was dissected and kept in Ringer at 0° C. The different regions of the brain (cerebrum, cerebellum and medulla oblongata) were separated with sterilized instruments, weighed in ice-cold Ringer and immediately used for analysis.

Proteins from the samples were precipitated by trichloro acetic acid (BDH) and estimated by the micro-biuret method[10].

RNA was extracted by the method of Schmidt-Thanhausser-Schneider[11] and estimated by orcinol colour reaction following the colorimetric procedure described by Glick[12].

The Aspartate aminotransferase (AAT, E.C. 2·6·1·1) and alanine amino transferase (AIAT, E.C. 2·6·1·2) activities were determined following the colorimetric procedure of Reitman and Frankel as described by Bergmeyer[13].

RESULTS AND DISCUSSION

Results are summarized in Tables I–III.

Brain weight and body weight decreased considerably in the thiamine-deficient state (Table I).

TABLE I

Body and Brain weight of normal (control) and thiamine-deficient chicken[1]

Body weight in g Control	Body weight in g Test	Brain weight in g Control	Brain weight in g Test
82·7 ± 11·0	31·3 ± 5·5	1·97 ± 0·2	1·37 ± 0·2
75·8–98·8*	29–40·8*	1·8–2·2*	1·1–1·5*

[1] Values are mean ± S.D. of 28 observations.
* Represents the weight range.
Test = Thiamine-deficient chicken.

Such a differential response of the different regions of the brain may be related to the differences in the functional status of the region concerned. However, the general increase in RNA and protein levels observed in the present investigation points to an augmentation in the activity of the protein synthetic machinery in B_1-hypovitaminous birds.

The activity levels of AAT and AlAT, in general, showed an increase in all the compartments of the brain studied, *viz.*, cerebrum, cerebellum and medulla oblongata during thiamine deficiency (Table III). It is also seen (Table III) that the increase in the levels of the aminotransferase activities showed variation in relation to the region of the brain. Further, the brain stem showed marked response for the changes in the activity levels of AAT and the cerebrum demonstrated the maximum response for changes in the activity levels of AlAT (Table III). It is also noteworthy that the activity level of AAT was more than that of AlAT in the CNS of the thiamine-deficient birds. The general increase in the activity levels of both the aminotransferases during thiamine deficiency may be either due to the activation of the enzyme synthesis by the altered cellular environment prevailing in the brain, or due to an increase in the rate of enzyme synthesis. Supporting this suggestion, an increase in the levels of RNA and proteins have been detected (Table II).

TABLE II

Changes occurring in the levels of RNA and protein in different regions of the brain of normal (control) and Thiamine-deficient chicken, Gallus domesticus[1]

	Cerebrum Control		Cerebrum Test	Cerebellum Control		Cerebellum Test	Medulla Control		Medulla Test
RNA mg/g	4.75 ± 1·1	[a] +86·3 $p >$ 0·01	8·85 ± 1·2	4·8 ± 1·3	[a] +233·4 $p >$ 0·01	16·0 ± 4·0	4·3 ± 1·2	[a] +88·2 $p >$ 0·01	8.0 ± 1·9
Protein mg/g	106 ± 4·8	[a] + 0·46 NS	106·5 ± 7·6	105·5 ± 17·9	[a] + 5·6 NS	111·5 ± 13·9	75·0 ± 6·8	[a] + 8·0 NS	81·0 ± 7·4

[1] Values are mean ± S.D. of 10 observations.
[a] Percentage change, signs (+) indicate an increase over controls.
NS = not significant.
Test = Thiamine-deficient birds.

In general, the protein content increased (negligible in the cerebrum and cerebellum and considerable in the brain stem) on inducing thiamine deficiency (Table II) Paralleling the increase in proteins, RNA levels also increased (significant) in all the regions investigated during B_1-hypovitaminosis. Further, marked differences in the protein and RNA contents of the cerebrum, cerebellum and medulla oblongata were noted (Table II). A remarkable feature was that the cerebrum demonstrated least response for changes in RNA and also protein in the thiamine deficient state. Maximum response for changes in RNA was exhibited by cerebellum and for protein levels was exhibited by the brain stem on inducing thiamine deficiency (Table II).

AAT and AlAT are of great clinical significance[14]. The levels of these aminotransferases in serum have been studied as an index for the diagnosis of disease-conditions like hepatic and cardiac infractions[15]. The liver aminotransferase activities have been known to increase under several pathological conditions[14,15]. In support of this the aminotransferase activities of the brain have shown a significant augmentation during

TABLE III

Aminotransferase activities in different regions of the Brain of normal (controls) and Thiamine-deficient chicken[1] Gallus domesticus

	AAT			AlAT		
	Cerebrum	Cerebellum	Medulla	Cerebrum	Cerebellum	Medulla
Controls	1·15[2] ± 0·06	0·92 ± 0·06	1·2 ± 0·076	1·02 ± 0·07	1·1 ± 0·06	1·34 ± 0·23
Test	8·7 ± 2·6	8·7 ± 1·5	11·6 ± 1·5	2·4 ± 0·05	1·95 ± 0·2	2·6 ± 0·08
	[a]+656·5	[a]+846·6	[a]+866·7	[a]+134·7	[b]+ 77·2	[a]+94·0
	$p >$ 0·01	$p >$ 0·01	$p >$ 0·01	$p >$ 0·05	$p >$ 0·01	$p >$ 0·05

[1] Activity expressed as micromoles of keto acid formed /g wet weight of the tissue/hr.

[2] Values are Mean ± S.D. of 12 observations.

[a] Percentage change, sign (+) indicating an increase over controls.

thiamine deficiency in chicken. Since the aminotransferases are known to link amino acid and carbohydrate metabolism[14], an increase in aminotransferase activities during thiamine deficiency indicates that the contribution of glucogenic amino acids to the carbohydrate metabolism is more. It is therefore possible that carbohydrate metabolism is not maintained at a steady level during thiamine deficiency. The lower activity levels of AlAT compared to the activity levels of AAT account for the decreased production of alanine from pyruvic acid in the brain of B_1-hypovitaminous birds. This is of significance in the metabolism of brain as alanine plays a vital role in linking carbohydrate and protein metabolism[14]. This is in accordance with the earlier reports of Peters[5] and Nalini and Nayeemunnisa[9].

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The author is thankful to the late Prof. K. Pampapathi Rao for facilities.

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1. Lajtha, A., *Handbook of Neurochemistry*, Vol. VII, Plenum Press, New York 1972.
2. —, *Regional Neurochemistry*, S. S. Kety and J. Elkes, eds., Pergamon Press, Oxford, 1961.
3. —, *Int. Rev. Neurobiol.*, 1964, **7**, 1.
4. —, *Ibid.*, 1964, **6**, 1.
5. Peters, R. A., *Biochemical Lesions and Lethal Synthesis*, Pergamon Edition, International Series of Monographs on Pure and Applied Biology, 1963, p. 8.
6. Richter, D., *Brit. Med. Bull.*, 1965, **21**, 76.
7. Lehr, P. and Gayet, J., *J. Neurochem.*, 1963, **10**, 169.
8. Richter, D., *Protides of the Biological Fluids* H. Peeters, ed., Vol. 13, 137–143, Elsevier, Amsterdam, 1965.
9. Nalini, U. P. and Nayeemunnisa, *Experientia*, 1976, **32** (2) 198.
10. Itzhaki, R. T. and Glick, D. M., *Analyt. Biochem.*, 1964, **9**, 401.
11. Schmidt–Thannhausser–Schneider (1957, as cited by Glick, *See* 12).
12. Glick, D., *Methods of Biochemical Analysis*, Interscience Publishers, New York 1964, Vol. 1.
13. Bergmeyer, H. U., *Methods of Enzymatic Analysis*, Academic Press, New York, 1965, p. 837.
14. Harper, H. A., *Review of Physiological Chemistry*, Maruzen Asian Edition, 1965.
15. Wroblewski, *Amer. J. Med.*, 1959, **27**, 911.
16. Chung and Kittong, *J. Cath. Med. Coll.*, 1970, **15**, 251.

LETTERS TO THE EDITOR

A DIRECT EVIDENCE FOR THE NON-EXISTENCE OF ESTER-BONDS IN COLLAGEN CHAINS

Introduction

THE sub-unit structure model of collagen chain with ester-bonds between the sub-units was earlier suggested by Gallop *et al.*[1,2], on the basis of their findings on the reaction of hydroxylamine and hydrazine with denatured collagen. This model was once considered as an acceptable one for collagen and the results in many laboratories were explained in view of this model. In an earlier communication[3] we have reported that the soluble fraction obtained by treating shark-fin rays (elastoidin) with formic acid consists of smaller sub-units having sedimentation coefficient ($S_{20,\omega}$) value of 1·6 compared to 3·2 for an intact single collagen chain. As formic acid under such experimental conditions does not affect peptide bonds[4-7], the observation was explained by assuming that shark-fin collagen chains are built up of smaller sub-units linked together by non-peptidic bonds, possibly of ester-type which might be broken by formic acid treatment. Possible mechanisms of formation of such ester-bonds *in vivo* in the growing polypeptide chain were also suggested[8,9]. But later, another group of workers[10-13], particularly Bornstein *et al.*[11-13] claimed that the sub-units which were obtained by treating collagen with hydroxylamine and hydrazine are not linked through ester-bonds but through cyclic imides formed by the condensation of the side-chain amide group of asparagine with amide nitrogen of the subsequent glycine residue in the chain. Both these groups, however, arrived at their own conclusions through indirect approaches. No direct proof for the presence or absence of ester-bonds in collagen chains has hetherto been offered. Considering that a study on the action of esterase on collagen chain should provide us, with a direct evidence regarding this, it was thought worthwhile to investigate the problem using a liver esterase as a probe. Liver esterases appear to have macromolecular substrate specificity also since these enzymes are considered to be reactive towards the ester-bond in the aminoacyl-*t*-RNA complex during the protein synthesis in liver[14].

Experimental

Preparation of Collagen Chain.—The swim-bladder removed immediately after slaughtering the carp was cleaned and cut into small pieces. These were then treated with 0·1% acetic acid for 24 h, after which the solution was filtered through glass wool. All operations were performed at 4° C. The filtrate was lyophilized and stored at 4° C in a desiccator. The α 1-chain was isolated from thermally (40° C for half an hour) denatured collagen by chromatography on CM-cellulose[15]. The desired fractions were pooled, dialysed and lyophilized.

Preparation and Purification of the Esterase.—Esterase was extracted from rat liver according to Okuda and Fuji[16] and treated with pancreatic lipase (Calbiochem) and purified by gel filtration on Sephadex G–200 (Pharmacia) as described by these authors.

Esterase Activity.—Two methods were employed to check esterase activity, a spectrophotometric method using *p*-nitrophenyl acetate (*p*NPA) as substrate[17] and a titrimetric method using methyl butyrate as substrate[18]. In the former method the enzyme prepared had a specific activity 320 ± 10 μ moles/mg protein/h at pH 7 and 25° C, whilst against methylbutyrate the specific activity was 250 ± 10 μ moles/mg protein/h at pH 8 and 25° C.

Proteolytic activity.—Proteolytic activity of the esterase was checked by using the method of Levisohn and Aronson[19]. To 12·5 mg portions of AZOCOLL (Calbiochem) suspended in 1 ml of 0·2 M sodium phosphate buffer (pH 7·6) 0·5 ml of the enzyme in the same buffer was added and the volume was brought to 2·5 ml with water. After incubation at 37° C for 15 minutes the suspension was filtered through Whatman No. 1 filter-paper. The absorbancy at 580 nM of the filtrate was measured in the spectrophotometer, Unicam, Sp-600.

Reaction of esterase with collagen.—Purified α 1-chain of Carp swim-bladder was digested with esterase at 25° C and also at 40° C for 15 hours at pH 8·0 with the collagen : esterase ratio 100 : 1. A pH-meter autotitrator was used to follow up the reaction.

Ultracentrifugation.—The sedimentation coefficient values of both enzyme-treated as well as untreated material were determined at 25° C using the Spinco-Model E (Beckman) ultracentrifuge. The concentrations of the protein in phosphate buffer ($pH = 7$, $\mu = 0{\cdot}1$) were kept the same (0·8%) in both cases.

Results and Discussion

Results summarised in Table I indicate that no hydrolysis of the collagen has occurred by treatment with the esterase. The $S_{20,\omega}$ value of 2·83 for the esterase-treated collagen was found to be almost the same as that (2·81) of the untreated one. These observations offer a direct evidence to support the view that no conventional ester-bonds are present in the collagen chain.

TABLE I

Reaction of esterase with different substrates and with collagen

	Reactions	*Remarks*
1. *Proteolytic substrates*		
AZOCOLL	No hydrolysis	No proteolytic activity present
2. *Ester substrates*		
p-Nitrophenyl acetate (*p*NPA) Methyl Butyrate	Hydrolysis	Esterase activity present
3. *Collagen*		
Purified α1-chain of Carp swim-bladder	No hydrolysis	Ester-bonds are not present in collagen chain

Our grateful thanks are due to Professor A. Sen, Bose Institute, for allowing us the use of some of his equipments.

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1. Gallop, P. M., Seifter, S. and Meilman, E., *Nature*, 1959, **183**, 1659.
2. —, *Ibid.*, 1966, **209**, 73.
3. Ghosh, S. K. and Saha, N. N., *Sci. and Cult.*, 1969, **35**, 432.
4. Josefsson, L. and Edman, P., *Biochim. Biophys. Acta*, 1957, **25**, 614.
5. Narita, K., *J. Am. Chem. Soc.*, 1959, **81**, 1751.
6. Smillie, L. B. and Neurath, H., *J. Biol. Chem.*, 1959, **234**, 355.
7. Ramachandran, L. K., *Biochem. Biophys. Res. Commun.*, 1962, **6**, 443.
8. Ghosh, S. K. and Saha, N. N., *Abstr. Third Intern. Biophys. Congress of the Intern. Union for Pure and Applied Biophys.*, Cambridge, Massachusetts, USA, IIF 1, 1969, p. 194.
9. Fahnenstock, S. and Rich, A., *Nature, New Biol.*, 1971, **229**, 8.
10. Butler, W. T., *J. Biol. Chem.*, 1969, **244**, 3415.
11. Bornstein, P., *Biochem. Biophys. Res. Commun.*, 1969, **36**, 957.
12. —, *Biochemistry*, 1970, **9**, 2408.
13. — and Balian, G., *J. Biol. Chem.*, 1970, **245**, 4954.
14. Goldberg, M. I. and Fruton, J. S., *Biochemistry*, 1969, **8**, 86.
15. Piez, K. A., Eigner, E. A. and Lewis. M. S., *Ibid.*, 1963, **2**, 58.
16. Okuda, H. and Fuji, S., *J. Biochem.* (Tokyo), 1968, **64**, 377.
17. Huggins, C. and Lapides, J., *J. Biol. Chem.*, 1947, **170**, 467.
18. Harrer, C. J. and King, C. G., *Ibid..* 1941, **138**, 111.
19. Levisohn, S. and Aronson, A. I., *J. Bacteriol.*, 1967, 1967, **93**, 1023.

A RELATION BETWEEN MAGNETIC SUSCEPTIBILITY AND ELECTRONIC POLARIZABILITY OF NOBLE GASES

THE molar diamagnetic susceptibility of a monatomic gas is given by the well-known expression

$$\chi_M = -\frac{Ne^2}{6\,mc^2}\sum_i \langle r_i^2 \rangle \tag{1}$$

where $\langle r_i^2 \rangle$ signifies the mean square radius for the i-th electron and the summation extends over all the electrons of the atom. Thus the theoretical calculation of diamagnetic susceptibility for any monatomic gas reduces to the evaluation of the integral

$$\langle r^2 \rangle = \int \psi^* \, r^2 \, d\, a\, \tau \tag{2}$$

where ϕ is the wave function for the normal state in zero field. Wave functions obtained by analytical and other approximations are usually employed in the calculation of χ_M, since, except for hydrogen, which is not diamagnetic, the wave functions are not known exactly.

Expression (1) shows that the value of χ_M is dependent essentially on the effective size of the charge distribution within the atom. When the charge distribution is spherically symmetric, one may assume that the magnitude of the susceptibility is determined by an effective charge distribution having an average radius. In this case the experimental values of diamagnetic susceptibility for monatomic gases may be considered as a measure of the size of the charge distribution, in so far as the response of the electron cloud to a magnetic field is concerned.

When the atom is subjected to an electrostatic field the charge distribution gets displaced with respect to the positively charged nucleus. The polarization effect depends on the effective volume of the electron cloud and hence, it must be related to the cube of the average radius of the charge distribution. It is reasonable to assume that the charge distribution which responds to the electric field will have similar characteristics as the one which determines χ_M. Thus in spherically symmetric atoms like the atoms of noble gases one may expect some simple relationship between the diamagnetic susceptibility and electronic polarizability.

In Fig. 1, the square root of the modulus of molar susceptibility[1], $|\chi_M|^{1/2}$, is plotted against the cube root of electronic polarizability[2], $\alpha^{1/3}$, for five noble gas atoms. Except in the case of neon the remaining four elements lie on a straight line.

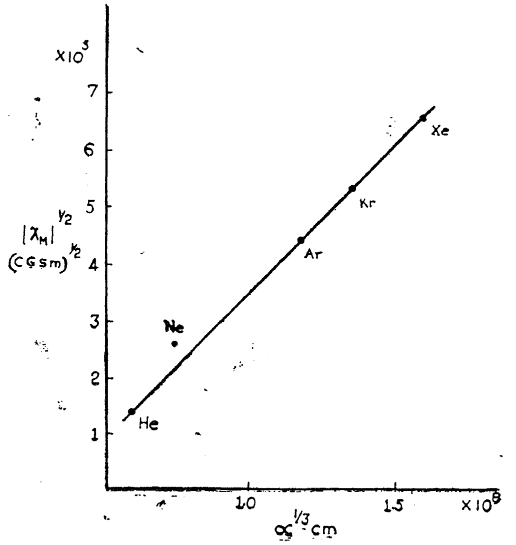

FIG. 1. Graph between square root of susceptibility and cube root of polarizability, for noble gases.

If the experimental value of susceptibility is taken as correct, then the graph suggests a value of 0·55 for the polarizability of neon, which is slightly higher than the present value of 0·40. A general relationship between molar susceptibility and polarizability may be expressed as follows :

$$|\chi_M|^{1/2} = a\,\alpha^{1/3} - b \qquad (3)$$

The constants a and b have numerical values 5·12 and 1·63 respectively when χ is in 10^{-6} cgsm units and α is expressed in units of 10^{-24} cm^3. This simple relationship and the graph given above are good indications of the common size effect of the extranuclear electron cloud that underlie both magnetostatic and electrostatic phenomena, *viz.*, diamagnetic susceptibility and electronic polarizability.

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Cochin 682 022, *October* 6, 1975.
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1. Weast, R. C., (ed.), *Hand-Book of Chemistry and Physics*, (55th edition), C. R. C. Press, Inc., Cleveland, 1974.
2. Rao. C. N. R., George, M. V., Mahanthy, J. and Narasimhan, P. T., *A Hand-Book of Chemistry and Physics*, Affiliated East-West Press, Pvt, Ltd., New Delhi, 1967.

FATTY ACID COMPOSITION OF TRIGLYCERIDES OF *ESCHERICHIA COLI* LIPIDS

Introduction

IT has been reported that *E. coli* phospholipids contain lauric, myristic, palmitic and stearic acids as saturated, palmitoleic and *cis*-vaccenic acids as unsaturated and *cis*-9, 10-methylenehexadecanoic and lactobacillic acids as cyclopropane acids[1-4]. But there is no report about the fatty acid composition of the triglyceride fraction of *E. coli* lipid. This communication deals with the fatty acid composition of *E. coli* triglycerides.

Materials and Methods

E. coli K 12 cells were grown in tryptone-yeast extract medium at 37°, log phase cells were harvested by centrifugation, washed with normal saline and kept at — 22°.

The lipids were extracted from the cells by the procedure of Bligh and Dyer[5] and fractionated on 100–200 mesh silicic acid columns (Mallinkrodt Chemical Works, St. Louis, Missouri) according to Vorbeck and Marinetti[6]. The triglycerides were isolated from the neutral lipid fraction by preparative thin-layer chromatography using the solvent system containing petroleum ether (40–60°), diethyl ether, acetic acid (80 : 20 : 1·5. v/v). The band having the same R_f as triolein was isolated and confirmed as triglyceride by pancreatic lypase hydrolysis[7] to yield 2-monoglycerides and free fatty acids.

Methyl esters of the component fatty acids were prepared directly from the triglycerides by methanolysis as described by Luddy *et al.*[8]. The methyl esters were analysed by vas-liquid chromatography (GLC). An "F and M Model 700–R" dual column analytical gas chromatograph was used for this purpose. Two 6 ft. × 0·25 in. columns one of 5% diethylene glycol succinate polyester (DEGS) and the other of 5% silicone gum rubber (SE 30) both on 60–80 mesh Gas-chrom Z (Applied Science Laboratories, State College, Pa.) were used. The former column was kept at 150° and the latter at 200° during operation.

The fatty acids were identified by comparing their retention times with those of authentic methyl esters and by chainlength determination according to Ackmann *et al.*[9], before and after hydrogenation. The position of double bonds in the mono-unsaturated fatty acids was determined by the permanganate periodate oxidation method of Von Rudloff as adopted to microscale by Scheuerbrandt and Bloch[10] followed by GLC identification of the resulting mono and dicarboxylic acids. The presence of cyclopropane fatty acids was confirmed according to Kaneshiro and Marr[11].

Results and Discussion

The triglycerides contained the following percentages of fatty acids; lauric (2·4), myristic (6·4), pentadecanoic (8·0), palmitic (22·8-, stearic (trace), palmitoleic (9·6), *cis*-vaccenic (25·6), *cis*-9, 10-methylene hexadecanoic (10·8) and lactobacillic (14·4). All these fatty acids have previously been reported to be present in *E. coli* phospholipids[1-4]. Several workers reported the 15-carbon acid to be a cyclopropane fatty acid[3] but during our investigation it was proved to be a straight chain, saturated fatty acid because it did not change its position on GLC on drastic hydrogenetion according to Kaneshiro and Marr[11].

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1. Bishop, D. G. and Work, E., *Biochem. J.*, 1965, 96, 567.
2. Kanemasa, Y., Akamatsu, Y. and Najima, S., *Biochim. Biophys. Acta*, 1967, 144, 382.
3. Cronman, J. E., Jr., *J. Bacteriol.*, 1968, 95, 2054.
4. Chattopadhyay, P. K. and Dutta, J., *Lipids*, 1975, 10, 497.
5. Bligh, E. G. and Dyer, W. J., *Can. J. Biochem. Physiol.*, 1959, 37, 911.
6. Vorbeck, M. L. and Marinetti, G. V., *J. Lipid. Res.*, 1965, 6, 3.
7. Mattson, F. H. and Volpenhein, R. A., *Ibid.*, 1961, 2, 58.
8. Luddy, F. E., Bardford, R. A., Herb, S. F. and Magidman, P., *JAOCS*, 1968, 45, 549.
9. Ackman, R. G. and Burgher, R. D., *Ibid.*, 1965, 42, 38.
10. Scheuerbrandt, G. and Bloch, K., *J. Biol. Chem.*, 1962, 237, 2064.
11. Kaneshiro, T. and Marr, A. G., *Ibid.*, 1961, 236, 2615.

NOTE ON THE EVOLUTION OF PHYTO-KARST TOPOGRAPHY IN THE GREAT LIMESTONE FORMATION OF JAMMU PROVINCE, J AND K STATE, INDIA

THE present note records for the first time the evolution of phytokarst (Folk R. L., *et al.*[4], 1973) topography in the Great Limestone formation of Jammu Province, J and K State. This peculiar landscape has been formed by subaerial endolithic algae, as a result the formation has been coated black with jagged pinnacles marked by an excellent delicate lacy dissection which do not exhibit any orientation. The phytokarst features have been noticed while making a sedimentological studies of the rock units between Muttal (Lat. 32° 59′ N: Long. 75° 01′ 30″ E) and Salal (Lat. 33° 10′ N: Long 74° 50′ E) (Figs. 1 and 2). The area lies

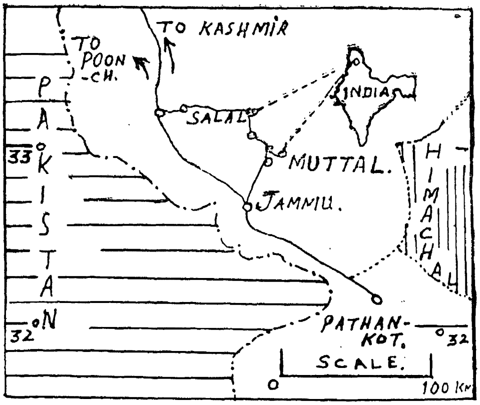

FIG. 1. Location Map.

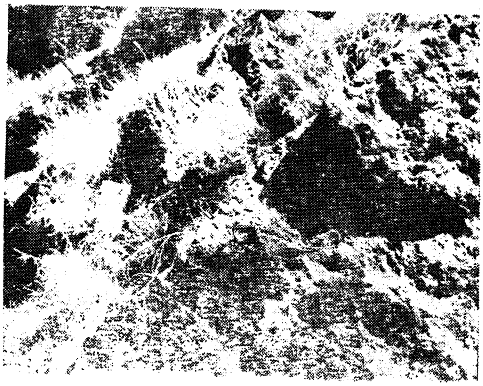

FIG. 2. Showing the development of phytokarst on the Limestone rocks.

on the Survey of India Topo Sheet Nos. 43 0/4, 43 P/1, 43 L/13 and 43 K/16 ; in the Udhampur District of Jammu. In this area, the Great Limestone is the oldest formation, exposed in the form of chain of inliers running in an echelon fashion. The limestone inliers are flanked on either side by Tertiary rocks of diverse characters. The Great Limestone is a massive formation whose base has not been observed anywhere in the area.

The area has received attention since 1876 but no work has been done on the geomorphology of the limestones. Sharma[8] (1970) while making a detailed study of the dolostones of the area made a reference to some Karst features but he did not study the phytokarsts. The phytokarst is a typical type of topography developed in the limestones by blue green algae, under sub-tropical climatic environments. In an effort to sculpture the limestones, the algae have been supported by rain, dew, etc. The biological activity gains strength with variations in the amount of temperature and rainfall. It is a rare rock sculpture.

The petrographic studies have shown that the phytokarst represents a highly dolomitised biolithites, with an extreme degree of diagenesis. The rocks are very compact with varying shades of colours. Most of limestones are coated black and dissected to make them a stony lacework resembling more or less to a sponge. At places, the limestone surface with black algal dots has tiny crystals of carbonate. The phytokarst topography has been noticed sparsely and not at a particular point on a large scale.

The phytokarst differs greatly from the normal Karst and the littoral Karst. The normal Karst is mostly caused by solution of inland carbonate rocks and do not have black coatings. The littoral Karst on the other hand, is the combined work of browsing and boring organisms and the solvent action of sea water. The area under study has already shown normal Karst features such as Karst windows, caves, clefts or clints and grykes but the occurrences of phytokarsts have been recorded for the first time. More detailed work in this direction is likely to throw more light on the subject and will be published soon.

My sincere thanks are due to my teacher Dr. T. R. Bhatia, whose inspiration and encouragement enabled me to do this work.

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Jammu, *October* 20, 1975.

---

1. Chadha, S. K., *Curr. Sci.*, 1975, 44, 665.
2. Cvijic Jovan, "The evolution of Lapies : A study in Karst physiography," *Geog. Rev.*, 1924, 14, 26.
3. Doerr, Arthur, H. and Hoy, Don, R., "Karst Landscapes of Cuba, Puerto Rico and Jamaica," *Sc., Monthly*, 1957, 85, 178.
4. Folk, Robert, L., *et al.*, *Bulletin. Geol. Soc. Am.*, 1973, 84 (7), 2351.
5. HodgKin, E. P., *Malaysia Geol. Soc.. Bull.*, 1970, 3, 27.
6. Jones, R. J., *Geol. Assoc. London, Proc.*, 1965, 76, 421.
7. Medlicott, H. B., *Recd. G.S.I.*, 1876, 9, 53.
8. Sharma, M. R., *Ph.D. Thesis*, University of Jammu, Jammu, 1970 (unpublished).
9. Sweeting, Marjorie, *Geog. Journ.*, 1958, 129, 184.
10. Wadia, D. N., *Mem. G.S.I.*, 1928, 51, 253.
11. —, *Ibid.*, 1937, 72, 162.
12. Wynne, A. B., *Ibid.*, 1872, 9 (2).

SHOCK AVOIDANCE LEARNING IN INTACT AND HEADLESS PREPARATIONS OF GRASSHOPPER *POECILOCERUS PICTUS*

Introduction

THE phenomenon of shock avoidance learning in orthopteran insects among invertebrates has been studied in greater detail recently[1-5]. Although a unit system that exhibits learning behaviour is important to a great extent in unfolding the mechanisms underlying the process of learning, the integrity of such a system in relation to the acquisition, storage and retention of learned behavioural information is of considerable interest. This paper is a comparative study of the learning behaviour in intact and headless systems of grasshopper *Poecilocerus pictus*.

Materials and Methods

Grasshoppers (*Species* : Poecilocerus pictus, *Group* : Exopterygota, *order* : Orthoptera, *Family* : Acrididae) were maintained in the laboratory in metal cages, on 12–12 hour light-dark cycle at 28 ± 1° C and 70 ± 10% relative humidity. They were fed *ad libitum* daily on the fresh leaves of milk-weed *Calotropis gigantea* (Linn.).

Two sets of adult male grasshoppers were selected for training. Each set consisted of 2 groups of 6 males each. In the first set, one group of insects were trained with their heads intact, while the other group of insects were decapitated before training. The second set of grasshoppers were injected with 0·02% prostigmin two hours before training and as in the first set, one group was trained headless and the other with heads intact. The severed neck region in headless animals was sealed with soft dental wax to prevent the loss of haemolymph. The shock avoidance learning procedure was based on the method originally described by Horridge[6]. The time taken for learning in normal and prostigmia-treated insects was recorded. All the trained insects were tested for the retention of learned behaviour individually using the original training procedure at different hours from the time of attaining the learning criterion.

Results

Observations on the learning pattern of headless and intact insects reveal that the headless insects respond more quickly to the training procedure as compared to the intact ones. The number of shocks received by the experimental insects during the initial five-minute period was considerably greater in intact animals as compared to the headless ones. The headless animals took almost the same time to learn as did the intact ones. (Table I). However insects treated with prostigmin—a parasympathetic stimulant, exhibited a tendency to learn faster than the normal insects and took less time to reach the learning criterion as compared to normal insects (Table I).

TABLE I

The mean average-time taken to reach the learning criterion in "Intact" and "Headless" preparations of (Male) grasshopper Poecilcerus pictus

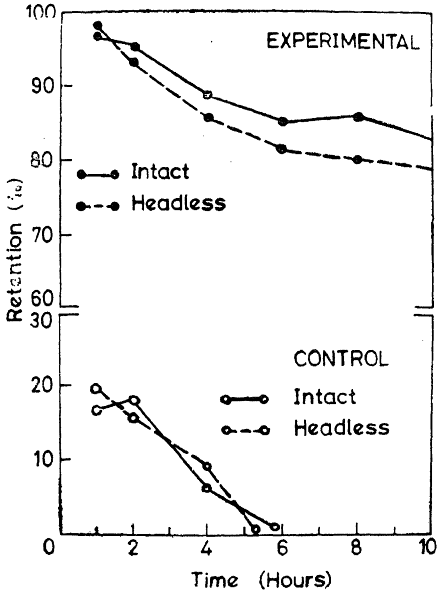
Experimental insect	Time taken in minutes*		t-values	P-values
	Normal	Prostigmin-treated		
Intact	41·8±3·9	31·9±7·6	3·7	<0·001
Headless	38·5±4·7	27·7±6·3	4·3	<0·001

(* Mean ± S.D. $n = 6$ for each type in each group of experimental insects).

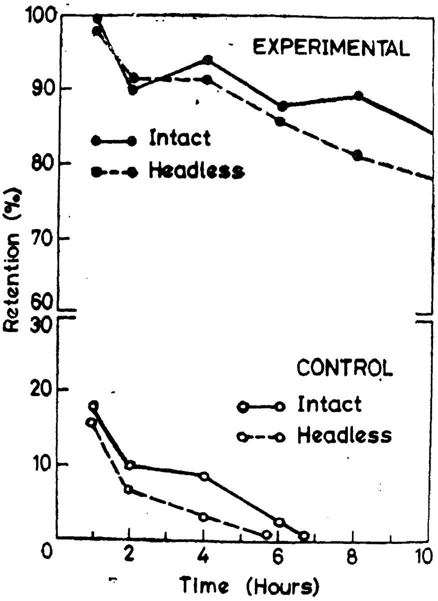
It is seen from the results of the retention test that intact experimental insect exhibits higher percentage of retention at the end of the observation period, while the headless experimental insect seems to reveal a comparatively low percentage of retention. On the other hand the prostigmin-treated insects appeared to show a noticeable consolidation of retention as compared to the non-treated experimental insects (Figs. 1 *a* and 1 *b*). The control animals in both groups did not learn the shock avoidance procedure and hence showed no significant retention.

Discussion

From the results it appears that the mode of learning as well as the retention pattern is dependent on the type of system used. The headless animals seem to respond immediately to the training process as compared to those with heads intact. This is also apparently evident from the results obtained on the prostigmin-treated insects. The extent to which the decapitation influences the learning process and its subsequent retention



(FIG. 1 *a*)



(FIG. 1 *b*)

FIGS. (1 *a*) and (1 *b*). *Retention Test*: The graph shows the percentage of retention in normal (1 *a*) and Prostigmin-Treated (1 *b*) insects during a post-training retention test.

depends perhaps not only on the immediate reaction mechanisms that follow the injury, but also on the relatively independent function of the ganglionic system that is capable of facilitating the acquisition of information at the segmental level. Although headless and intact systems do not differ significantly with regard to the acquisition of learning, the better performance of the headless animals during the initial five-minute period could be attributed to the integral organization of segmental ganglionic system that can independently regulate the acquisition of information irrespective of the imposed cephalic inhibition. It is probable that effective coordination between brain and nervous system in intact animals facilitates the retention capacity of the information processing system, although brain by itself may not directly influence the retention of acquired information. Inspite of the postural learning of leg position being predominantly a segmental phenomenon, a possible secondary role of brain in processing, storage and consolidating the retention of information could be considered to some extent in view of the transfer of learning known to occur between the segments of nervous system[7]. This aspect of study may throw some light upon the intermediary mechanisms involved in the process of learning and consolidation of traces of memory.

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---

1. Horridge, G. A., *Nature* (*London*), 1962 *a*, 193, 698.
2. —, In : *Learning and Associated Phenomena in Invertebrates*, (Eds., W. H. Thorpe and D. Davenport), Anim. Behar. Suppliment I, 1965.
3. Ebeling, W., Wagner, R. E., and Rierson, D. A., *J. Econ. Entomol.*, 1966, 59, 1374.
4. Kerkut, G. A., Oliver, G. W. O., Rick, J. T. and Walker, R. J., *Nature* (*London*), 1970 *a*, 227, 722.
5. Rick, J. T., Oliver, G. W. O. and Kerkut, G. A., *Q. J. Exptl. Psychol.*, 1972, 24, 282.
6. Horridge, G. A., *Proc. Roy. Soc.* (*London*), 1962 *b*. 157 B, 33.
7. Oliver, G. W. O., In : *Macromolecules and Behaviour*. (Eds. G. B. Ansell and P. B. Bradley), Macmillan, London, 1973, p. 113.

ON A NEW COCCIDIUM, *EIMERIA LOPHURAE* N.SP. (PROTOZOA : EIMERIIDAE) FROM KALIZ-PHEASANT, *LOPHURA LEUCOMELANA*

FROM pheasants, seven eimerian species have been described[1,3-5]. So far there is no report of any coccidium from Kaliz-pheasant, *Lophura leucomelana*. The persent communication deals with a new species, *E. lophurae* from this host.

Fresh droppings of a Kaliz-pheasant, collected during our visit to Delhi Zoo in December, 1974, revealed on microscopic examination coccidian oocysts. The material was left in 2·5% potassium dichromate solution for sporulation in a clean Petri-dish. Sheather's sugar solution was used for concentrating the oocysts by centrifugation[2].

Eimeria lophurae n. sp. (Fig. 1).

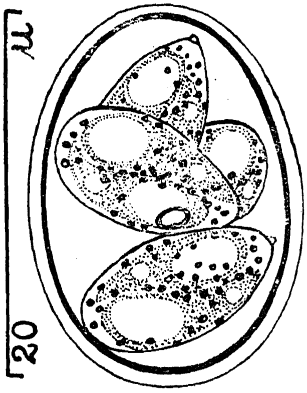

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FIG. 1. Sporulated oocyst of *Eimeria lophurae* n. sp.

Oocysts ellipsoidal, 20–24 μ × 14·6–17·3 μ (21·8 × 16·1) in size, had an oocystic wall of 1·3 μ thickness with an outer yellowish green and inner bluish brown layers. Length width ratio was 1·23–1·50 (1·35). Oocystic residuum absent. An ovoid dark polar granule present. Sporocysts, ellipsoidal with a narrower anterior end carrying a small knob-like stieda body, measured 10·7–12 μ × 5·3–6·7 μ (11·4 × 6·3). Sporocystic residuum present as dark scattered granules . Sporozoites, elongate, with one end broad and rounded and the other tapering carried one large and the other small refractile globule.

The present material differs from *E. dispersa* in which the larger oocysts have neither a polar granule nor a stieda body. In *E. phasiani* the oocysts are elongated ellipsoidal in shape, having a bright opercle and sporozoites carrying one globule. The oocysts in *E. pacifica*, *E. duodenalis* and *E. pictus* differ because of their ovoid shape. The oocystic shape and a greater oocystic size and thickness of its wall differentiate the oocysts of *E. megalostomata* from the present species. The large sized oocysts in *E. colchici* are easily distinguished by their elongated shape and the presence of a micropyle. The present oocysts are, therefore, assigned to a new species, *E. lophurae*—the first coccidium from Kaliz-pheasant (*Lophura leucomelana*).

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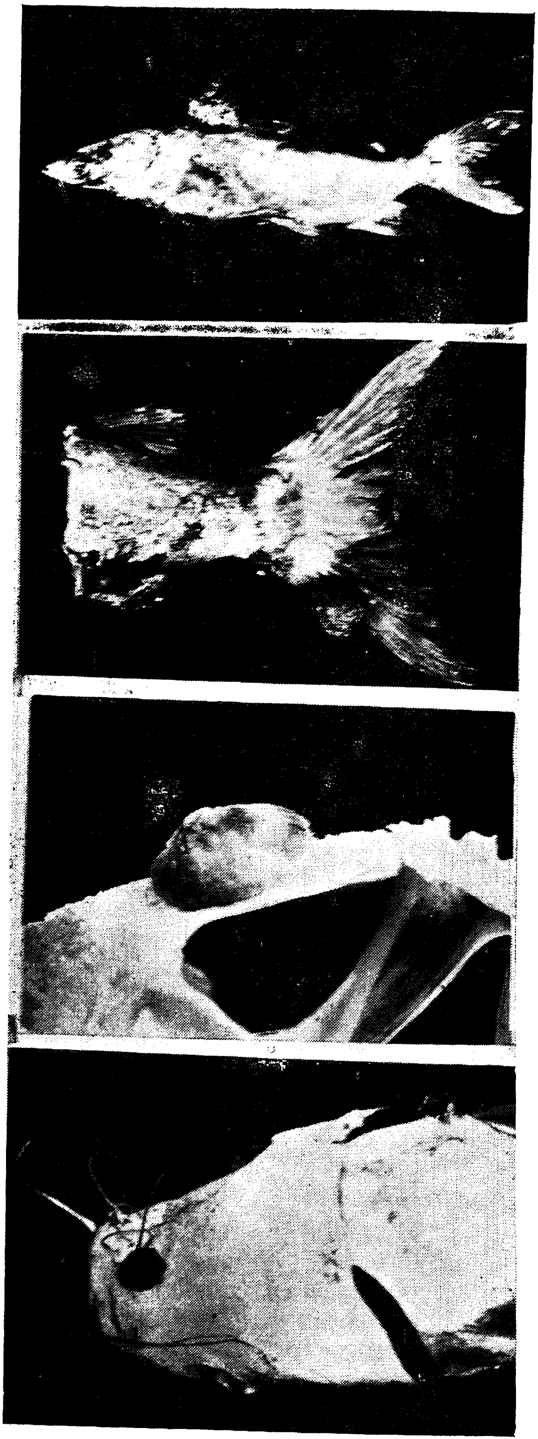
1. Bhatia, B. B., *Indian J. Microbiol.*, 1968, **8**, 193.
2. Levine, Norman, D., *Protozoan Parasites of Domestic Animals and of Man.*, Burgess Publishing Co., Minneapolis, Minnesota, 1961.
3. Norton, C. C., *Parasitol.*, 1967 *a*, **57**, 31.
4. —, *J. Protozool.*, 1967 *b*, **14**, 772.
5. Pellérdy. L., *Coccidia and Coccidiosis*, Akadémiai Kaidó, Budapest, 1965.

ON THE OCCURRENCE OF OSTEOMA IN A MARINE CAT FISH

TUMOURS of bone (osteoma) in fishes were first reported as early as the 17th century[1] and the latest is by Selvaraj *et al.*[2]. The occurrence of osteoma in the marine cat fish, *Arius maculatus* (Thunberg, 1792)[3] occurring in Porto Novo waters in the East Coast of India is reported here. A survey was made during the past 3 years (1973–1975) and interestingly *A. maculatus* was the only fish affected by osteoma. The details on number of specimens examined and the nature of tumours are given in Table I.

A total of 17 tumour affected fishes were collected during the period of survey. The caudal and dorsal fins were found to be the most commonly affected areas (Figs. 1 and 2). In 2 cases the tumours were found on the rostrum (Fig. 3—skin removed). In only one fish a tumour was found on one of the barbels (Fig. 4). The tumours



FIGS. 1–4.

TABLE I

Details of tumours occurring in the fish

Sl. No.	Date of collection	No. of specimens	Total length (mm)	Region affected	Size of tumour (Ht. × Dia. in mm)
1.	2–10–1973	1	580	Dorsal fin	10×14
2.	5–10–1973	2	(i) 476	Dorsal fin spire and rays scattered	5×14
			(ii) 512	Caudal fin rays	5×10
3.	20–10–1973	3	(i) 498	Base of the pectoral fin	7×11
			(ii) 441	Rostrum	5×17
				Caudal fin	5×9
			(iii) 530	Caudal fin scattered	3×12
4.	30–10–1973	1	462	Rostrum	5×15
5.	6–11–1973	1	570	Caudal fin	5×14
6.	10–11–1973	1	441	Dorsal rays	8×22
7.	11–11–1973	1	447	Infront of the first spine of the dorsal fin	40×45
				Dorsal fin rays	10×5
				Caudal fin	5×25
8.	17–11–1973	1	453	Caudal fin	20×15
9.	25–11–1973	1	466	Pelvic fin	17×9
10.	4–12–1973	1	400	Left side of the dorsal fin	15×10
				At opercular region	24×15
11.	18– 3–1974	1	382	Dorsal fin	5×15
				Pelvic fin	5×10·2
12.	1– 6–1974	1	432	Dorsal fin	3×8
				Pectoral fin	3·5×12·0
				Caudal fin	5×15·2
13.	1–11–1974	1	493	Caudal fin	7×17·8
14.	10–10–1975	1	482	Barbel	15×10

varied greatly in size from a pin head to 40 × 45 mm. These tumours occurred either singly or in groups of 3 to 5 in different sizes. The tumours with rough surface, were irregular in shape and had their origin in the bone. When the tumours were sectioned it was found to be a solid bony outgrowth with no space inside. Considering the total landing of *A. maculatus* at Porto Novo, the percentage of incidence of osteoma comes to only 2 to 7%. Recently Selvaraj *et al.*[2], reported on the occurrence of similar osteoma in 3 specimens of *T. jella* collected at South-West Coast of India. They observed osteochondroma in those specimens and this type was not observed in any of our specimens. Benign tumours of bone have been reported in 15 species of fishes and most of the tumours were probably sporadic occurrences[4]. The present report however indicates that there is a possibility of one species being more susceptible to bone tumours. These tumours in all probability represent exostoses which may be associated with a genetic predisposition in this species. Many attempts have been made to elucidate the etiology of such tumours but the exact reason is not known. A detailed knowledge of the epidemiology, pathology and cell biology of piscino neoplasm will not only help in ensuring the protection of fish populations but more significantly will serve to identify the presence of carcinogenic factors in the environment if any and their nature and mechanism of action.

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---

1. Mawdesley-Thomas, L. E., *Diseases of Fish*, (Ed.), Academic Press, N.Y., 1972, p. 191.
2. Selvaraj, G. S. D., Gopakumar, K. and Rajagopalan, M., *J.M.B.A.*, India, 1973, 15, (In press).
3. Fischer, W. and Whitehead, P. J. P., *Rome, FAO, Pag. Var. FAO Species Identification Sheets for Fishery Purposes, Eastern Indian Ocean (Fishing Area 57) and Western Central Pacific (Fishing Area 71)*, 1974, 1.
4. Scarpelli, D. G., In : *Fish in Research*, O. W. Neuhaus and J. E. Halver, Ed., Academic Press, N.Y., 1969, p. 45.

MAMMALIAN TESTIS IN ORGAN CULTURE

THE differentiating germ cell in the mammalian testis offers a good model for the study of mechanisms involved in selective gene activation and repression. The male gonad, however, is not amenable to experimental manipulations, since there are several complicating factors in *in vivo* systems which do not allow a proper evaluation of results[7]. Thus an *in vitro* system offers considerable advantages. Several attempts at cultivating the mammalian testis in organ culture have not produced encouraging results[1-3]. The present series of experiments deal with the intervals during meiotic differentiation which are particularly sensitive to explantation. Several techniques have been tried to cultivate the mammalian testis in organ culture and a comparison has been drawn between them. A new technique was tried which gave much better maintenance and possibly some differentiation.

Materials and Methods

Explantation experiments.—Ten adult male shrews were given an intraperitoneal injection of ^{3}H-thymidine and later sacrificed at 6 hr, 24 hr, 4 days, 7 days and 12 days post labelling. Two animals were used for each post labelling interval. Testes were removed aseptically, washed several times in Puck's saline (Mammalian) containing antibiotics. The *tunica albuginea* was carefully removed without disturbing either the seminiferous tubules or the interstitial cells. Each testis was then sliced into two or three parts each measuring about 2 mm^3. These were then placed side by side on a stainless steel wire grid, covered with teabag paper enclosed in a large sized Leighton tube. Mammalian Tissue culture medium, TC 199 (Difco) fortified with 15% foetal calf serum (GIBCO) antibiotic (100 IU/ml. penicillin, 100 mg/ml, streptomycin, 2·5 μgm/ml Fungizone) was then added so that the medium just reached the lower surface of the teabag paper and thus wetted it. The explants thus remained at the gas-liquid interphase. The Leighton tube was stoppered airtight to prevent change in pH (Table I gives the duration, each explant was in culture).

Comparative study of culture techniques.—Table II gives the various techniques that were tried for the culture of mammalian testis in organ culture. The general procedure was the same in all the techniques, and they differ only in the constituents of the organ culture medium except where otherwise mentioned.

TABLE I

Showing result of explantation experiments

Exp. No.	Labelled cells at the time of explant	Days after explantation	Labelled cells after explantation	Remarks
I. (*a*)	Premeiotic Interphase	5	Zygotene	*Sertoli spermatogonia*, Early pachytene absent, late pachytene pycnotic.
(*b*)	,,	22	Nil	Sertoli present, large nucleated gonocytes present.
II. (*a*)	Zygotene	4	Nil	*Early pachytene absent*, middle and late pachytene pycnotic, large nucleated gonocytes present.
(*b*)	,,	10	Nil	*Premeiotic interphase* and early pachytene absent. Mid and late pachytene pycnotic.
III. (*a*)	Leptotene	2	Zygotene	Large nucleated gonocytes present.
(*b*)	,,	15	Nil	Large nucleated gonocytes present.
IV.	Mid-pachytene	15	Nil	Large nucleated gonocytes present.

TABLE II
Showing the details of five culture technique

Sl. No.	Culture medium	3–H–TdR added on	Samples taken on/after explantation	Incubation temperature	Remarks
I.	T.C. 199–Antibiotics + Fungizone Foetal Calf Serum 15%	At the time of setting culture	2 days, 3 days and 7 days	37° C	Tubules were suspended in the medium, taken in a blood culture bottle. Air dry preparations were made.
II.	Eagle MEM+Antibiotics + Fungizone + Foetal Calf serum 15% + L-glutamine 292 mg/litre + testosterone 50/μ gm/ml.	..	2 days, 4 days and 6 days	31–33° C	Four days rats were used. Petri-dishes with steel grids covered with teabag paper was used. 8/μ sections were cut on Bouin's or neutral buffered formalin fixed tissue.
III.	T.C. 199 + Antibiotics + Fungizone + Foetal Calf serum 15% + FSH (100/μ g/ml)	..	8 days	31–33° C	Large sized Leighton tubes with stainless steel grids covered with teabag paper.
IV.	T.C. 199 + Antibiotics + Fungizone + Foetal Calf serum 15% + FSR (100/μ gm/30 ml) NIH 5–8 HCG (10/μg/30 ml)	13 days before explantation	15 days	31–33° C	Same as above
V.	Eagle MEM + Antibiotics + Fungizone + Foetal Calf Serum 15% + FSH–NIH S-8 (50/gμm/ml) + LH (10 IU/ml)	7 days after explantation	4 days, 7 days after 3H–TdR labelling	31–33° C	Same as above

Observations

Table I. shows the results of the explantation experiments. It is clear that the meiocyte, most sensitive to explantation is the pachytene (Mid and Late), which stages of meiosis are metabolically very active. The interval Zygotene-pachytene seems to be much more sensitive than Pm I-Leptotene-Zygotene (Experiment I). Mid pachytene did not develop to metaphase I. At later intervals, premeiotic interphases, leptotene and Zygotene, also disappear in the culture; the cell types present, even after prolonged periods of culture are the sertoli cells, and cells with large nuclei, which resemble the primitive genocytes, some of them in mitosis, spermatids (Spade type) and sperm. The cell types present in explants cultured for long intervals (three weeks) resemble to a very large extent, those present in the testis of hypophysectomized animals.

In all cases, a gradual loss of meiocytes occurred within 10 days of explantation. Of the several technique tried, none except V, proved to be an improvement. With the last technique, good maintenance of the explants was observed upto 14 days in culture (Fig. 1). Autoradiography was performed only on samples taken 4 days after 3 H–TdR administration, and it showed labelled Zygotene spermatocytes.

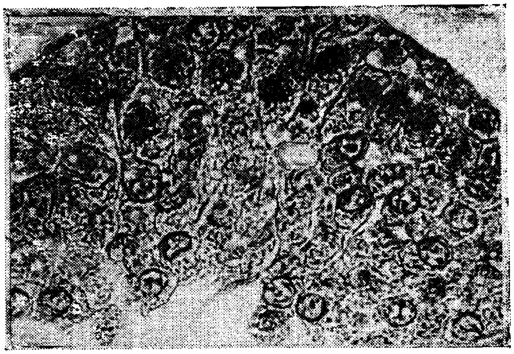

FIG. 1 *a*. Testicular explant cultured in the modified technique for 14 days, showing well maintained spermatogonia, spermatocytes and spermatids, × 1,250.

Discussion

The presence of histologically normal sertoli cells, primitive gonocytes in sitosis and spermatida (Spade type) in the cultured testis shows, that the culture media, and other conditions of culture are conducive for the maintenance of cells not undergoing differentiation. Therefore, the gradual elimination of the differentiating cells (Spermatogonia and spermatocytes) shows that every stage of this differentiation process is under the control of exogenous substances (hormones). This is, not withstanding, the appearance of labelled zygotene after 5 days in culture, when initially the label was present only in the premeiotic interphase. This can be attributed to residual hormonal activity in the explants. This conclusion does not agree with that of Steinberger and Steinberger[3], who have shown in their schematic diagram several intervals in the male germ cell development, which are independent of hormonal control.

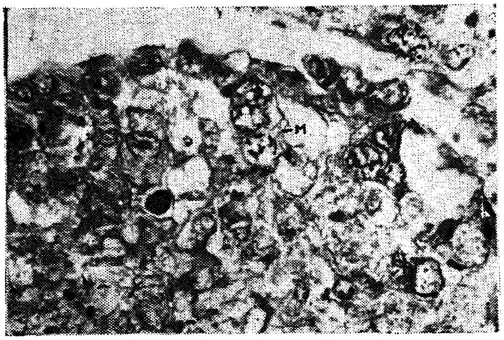

FIG. 1 *b*. Same as above, showing a spermatogonium in metaphase, × 750.

Several attempts at culturing the mammalian testes have not been totally successful. The explantation studies of Stern and Hotta[4] in *Trillium erectum*, provide a comparison. These authors have shown four distinct intervals during which meiotic development is susceptible to explantation. Three such intervals are during the premeiotic 'S'. The other interval extends between the end of Zygotene and Pachytene. Explantation at these intervals results either in reversion to mitosis of abnormal meiotic development. The present study shows that in mammalian systems, reversion of meiocytes to a mitotic division does not occur, but the meiocytes undergo degeneration. However, prolonged cultivation (12 days or more) in hormone free culture medium results in the appearance of large cells (gonocytes ?) undergoing mitosis. Steinberger and Steinberger[5] have obtained very encouraging results using L-glutamine, Vitamin A, E and C. They find that the addition of exogenous hormones (FSH, HCG and Testosterone) does not initiate or maintain spermatogenesis *in vitro*. The present study, however, shows that FSH-(NIH S-8) and LH (NIH) in combination can bring out a better maintenance, since viable premeiotic interphases were present even after 7 days in culture, which could incorporate 3 H-TdR and develop into Zygotene in 4 days, a time, period same as in *in vitro* conditions[6]. This may be due to the fact that while explanting the testicular fragments, we take precautions to keep the interstitial elements intact and culture all the pieces together. The intact interstitial cells can therefore respond to the action of gonadotrophins, and result in the production of endogenous androgen, which results in better maintenance. Absence of response to pituitary gonadotrophins *in vitro* reported earlier[3] might be due to the absence of interstitial cells as a result of loss during explantation.

The role of androgens in initiating meiotic development has been established[3] however, the exact mechanism is not known. Some of our experiments which are in progress suggest that low dosages of testosterone acting over a long time can result in slowing down of DNA synthetic rate and an increase in the DNA synthetic period, extending upto the meiotic prophase. This in turn increases the possibility of unreplicated regions of DNA coming together by chance collission some of which may result in pairing.

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1. Steinberger, A. and Steinberger, E., *J. Reprod. Ferti.*, 1965, 9, 243.
2. — and —, *Ibid.* (supplement), 1967, 2, 117.
3. Steinberger, E. and Steinberger, A., *The Gonads, Biochemical Endocrinology Monograph*. Ed., K. W. McKern, Appleton—Century—Crofts, 1969.
4. Stern, H. and Hotta, Y., "Symposium of nuclear physiology and differentiation," *Heredity, (supplement)*, 1969, 61 (1), 27.
5. Steinberger, A. and Steinberger, K., *Exptl. Cell. Res.*, 1966, 44, 429.
6. Rao, S. G. A., *Ph.D. Thesis*, Delhi University, 1974.
7. Steinberger, E. and Steinberger, A., *The Spermatogenic Function of the Testes, the Gonads*, Ed., K. W. McKerns, North-Holland, 1969, p. 715.

IMPROVEMENT OF SCENTED RICE VARIETIES (BASMATI 370) THROUGH INDUCED MUTATION

AT the Central Rice Research Institute, a breeding programme was initiated in 1971 to improve the yielding ability and arrest the lodging habit of fine and scented rice varieties through both conventional and mutation breeding. Significant results obtained through mutation breeding in the improvement of one such variety. Basmati 370, retaining all its original grain quality characteristics are reported in this paper.

Dry seeds of Basmati 370 (10% moisture) were treated with 20 Kr gamma rays from 60_{co} source at IARI during June 1971. M_1, M_2 and M_3 generations of these irradiated seeds were grown during subsequent seasons. Most of the morphological mutants showing marked changes along with culm shortening were screened out and were grown separately. Short statured mutants with no change in grain and cooking qualities from that of the parent were retained. In M_4 generation, 16 promising semidwarf mutants were finally isolated. These mutants bred true in M_5 generation and were assessed for yielding ability, grain size, appearance, kernel elongation and scent in M_5 and M_6 generations.

Data presented in Table I showed that the mutants of Basmati 370 were shorter in height and had more ear bearing tillers and had given higher yield (33–133%) than the control. At the same time, there was no alteration either in their duration, grain appearance, cooking quality or aroma,. It was noted that some of the mutants had better kernel elongation on cooking than that of Basmati 370.

In the earlier attempt to induce high yielding semidwarf mutants in scented varieties[5], the mutants reported seemed to have lost their grain quality or proved lower yielding than the parent. Reddy and Reddy[7] obtained promising dwarf mutants from HR. 47 (Basmati) which were earlier and yielded (70%) more than the control. Likewise, dwarf mutants had been isolated in the scented varieties NP. 49 (Dehra Dun Basmati) and NP. 114 (Kamod, scented) with a fair degree of resistance to blast, bacterial leaf blight and tungro virus[4] but these were found to be inferior to high yielding semidwarf varieties. At the Central Rice Research Institute, Misra (unpublished) isolated a semidwarf high tillering mutant in the short grained scented rice variety T. 412 without scent.

Scent is a heritable character and its presence has been reported to be governed by two to three pairs of genes[6]. Grain size and shape are governed by polygenic systems[1] and hence it is a tardy process to fix such characters through recombination breeding. Introduction of an alien dwarfing gene seemed to disturb the genic alignment controlling scent and cooking quality and to render identification of scented segregants in rice varieties through recombination breeding more difficult. On the contrary improvement in cooking quality and other grain characteristics has been feasible through induced mutation[2,3]. These scented semidwarf

TABLE I

Agronomic characters of Basmati 370 *mutants*

Mutant No.	Height (cm)	Average ears (No.)	Elongation ratio on cooking	Computed yield potential (kg/ha)	% yield over Basmati control
5712–4	95	28	2·28	4158	133
5713–10	100	24	2·26	3670	106
5713–14	90	19	2·24	3148	76
5712–8	99	20	2·22	2970	66
5707–1	94	18	2·16	2970	66
5709–5	93	21	2·14	2768	55
5708–5	106	19	2·12	2376	83
5713–7	89	20	2·10	2376	83
Basmati–370 (Control)	132	14	1·62	1782	..

mutants, because of their higher yield potential, could be grown as commercial varieties or used as scented semidwarf donors for improvement of other scented rice varieties. Thus, when the breeding objective is elimination or modification of one or two characters with the integrity of all other characters in tact in a rice cultivar, mutation breeding seems to be a more elegant and quick approach.

Thanks are due to the Director, Dr. S. Y. Padmanabhan, for his keen interest in this work.

Division of Genetics, C. GANGADHARAN.
Central Rice Research Institute, R. N. MISRA.
Cuttack-6 (Orissa), A. K. GHOSH.
December 17, 1975.

1. Ghose, R. L. M., Ghatge, M. S. and Subramanyan, V., *Rice in India,* 2nd Ed., ICAR, New Delhi, 1960.
2. Govindaswami, S., Ghosh, A. K. and Misra, R. N., *Indian J. Agric. Sci.,* 1972, 42 (10), 869.
3. —, — and —, *Proc. Symp. Use of Radiation and Radio Isotopes in Studies of Plant Productivity,* Pantnagar, India, 1973. p. 53.
4. Mahabal Ram, *Proc. SABRAO 2nd Gen. Cong.,* New Delhi, India (Abstract), 1973, p. 77.
5. Miah, A. J. and Bhatti, I. M., *IAEA Tech. Rep. Ser.,* No. 86, 1968, p. 75.
6. Nagaraju, M., Choudhury, D. and Balakrishna Rao, M. J., *Curr. Sci.,* 1975, 44, 599.
7. Reddy, G. M. and Reddy, T. P., *Proc. Symp. Use of Isotopes and Radiation in Agric. and Animal Husb.,* 1971, p. 237.

A PARTIALLY SELF-COMPATIBLE SUNFLOWER

THE low seed yields of sunflower (*Helianthus annuus* L.) are due to the occurrence of a high degree of unfilled or empty seeds ranging from 10 to 50%[1]. The problem is further accentuated by the sporophytic self-incompatibility system[2] which prevents seed set under self-pollination. The high degree of empty seededness is a major genetical problem[1] and therefore the incorporation of certain degree of self-compatibility would ensure seed filling eliminating the need for pollinating agents. Some of the lines and hybrids developed were found to be partially compatible[3,4]. However, the degree of self-compatibility was not reported. The discovery of a spontaneously occurring self-compatible sunflower line and its performance under selfing and in crosses is reported here.

The self-compatible (SC) mutant has a distinct plant type with conical appearance and additional inflorescences in the terminal axils of leaves (Fig. 1). The progeny derived from this exhibited wide variability for several morphological characters. The plants were of different heights (200–250 cm) and later in duration (120–140 days). The leaves numbering 80 to 98 were typically light green in colour. The diameter of the head ranged from 9 to 19 cm at maturity. The seeds (achenes) were small with thick ashy white hull and small kernels.

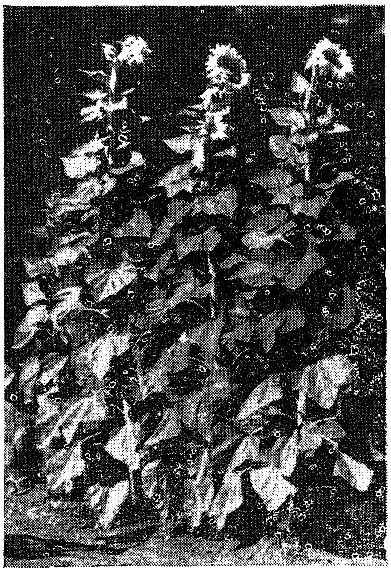

FIG. 1. Partially compatible sunflower plants.

TABLE I

Seed set under selfing in sunflower

Particulars	Range %	Mean
A. Self-incompatible (SI)		
EC 27501 (10)*	0·0– 0·0	0·0
Sunrise (13)	0·0–46·7	8·3
EC 77195 (12)	0·0–38·7	8·5
B. Self-compatible (SC)		
(Plant numbers)		
289 (2)	13·0–56·9	35·0
290 (3)	4·5–37·0	25·8
291 (8)	0·0–54·3	30·5
292 (2)	42·8–69·9	56·3
293 (11)	25·5–85·8	54·0
C. Hybrids (F_1's)		
SC × EC 77195 (7)	0·0–64·0	26·6
SC × EC 27501 (13)	0·0–82·9	41·6
SC × Unknown (14)	0·0–80·8	59·3
SC × Sunrise (1)	0·0–80·8	65·1
D. Group means		
SI	0·0–46·7	5·6
SC	0·0–85·8	40·3
F_1's	0·0–82·9	48·2

* Number of plants.

The filled seeds in the self-compatible mutant ranged from 25 to 56% as compared to less than 9% observed in the self-incompatible types in the present studies (Table I) and elsewhere[5]. Some of the self-compatible plants were crossed with selected self-incompatible types without emasculation. The true hybrids were identified and data collected. The improvement in the seed set achieved by utilising the self-compatible mutant is given in Table I. The filled seeds in the F_1 hybrids ranged from zero to 83% with an average mean of 48·2%. Thus there is considerable scope for isolating self-compatible lines from the segregating material. It is also possible that the percentage of filled seeds in these partially compatible hybrids or lines could substantially increase under open-pollination. Apart from mutational rectification of excessive vegetative growth, duration and seed characters, the genetics of self-compatibility is also being investigated.

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December 29, 1975.

G. Harinarayana.
Miss P. Kamalam.
N. Ganga Prasada Rao.

1. Rajan, S. S., *Indian J. Genet.*, 1974, **34 A**, 669.
2. Stojanova, J., *Genetika i Selekcija*, Sofia, 1969, 2, 265; *cf. PBA*, 1970, 40, 8230.
3. Shuster, W., *Angew. Bot.*, 1970, 44, 87.
4. Fick, G. N., Zimmer, D. E. and Kinman, M. L., *Crop. Sci.*, 1974, 14, 912.
5. Skoric, D., *Savremen. Poljopr.*, Novisod, 1969, 17, 131; *cf. PBA*, 1970, 40, 8229.

JUVENILE HORMONE EFFECTS ON THE PREPUPAL AND PUPAL STAGES OF THE TOBACCO CUTWORM, *SPODOPTERA LITURA* FABRICIUS (NOCTUIDAE: LEPIDOPTERA)

Introduction

The use of analogues and antigonists of insect growth regulators, such as juvenile hormones and ecdysones for the suppression of insect populations is getting into vogue. 'Altozar' an analogue of insect hormone has been used in this work to determine its effect on the prepupal and pupal stages of *Spodoptera litura* Fab. in different concentrations.

Materials and Methods

The test insect culture was developed and maintained in the laboratory from a single egg mass collected from the cabbage fields around Hebbal (Bangalore). The larvae were reared on caster leaves throughout the insect life-cycle.

The juvenile hormone analogue (JHA)—"Altozar" was obtained from M/s. Zoecon Corporation, Palo Alto, California.

Concentrations of 0·5%, 0·75% and 1·0% of the JHA were prepared in 1% acetone solution; for control only 1% acetone was used. Each of these concentrations was applied to the prepupae individually on the dorsal surface using an automatic micro applicator at 5/μl per insect. The same concentrations were applied to the pupae topically at 1 μ/individual on the dorsal surface. In each experiment (Pre-pupae pupae) and for each concentration a unit of ten individual insects was used. Each set of experiments was replicated thrice. The experimental data are presented in Tables I and II.

Table I

Effect of Altozar on the prepupae of Spodoptera litura *Fabricius**

Sl. No.	Con. Altozar %	% of prepupal moulted into normal pupal	% of prepupal malformed or dead	% of adult emergence
1.	0·50	60	40	20
2.	0·75	30	70	20
3.	1·00	10	90	..
4.	Control	90	10	60

* Averages of three replicate experiments.

Table II

Effect of Altozar on the pupal of Spodoptera litura *Fabricius**

Sl. No.	Con. Altozar %	% of normal adult emergence	% of adults with malformed characters
1.	0·50	13·3	..
2.	0·75	20	6·6
3.	1·00	20	13·3
4.	Control	60	..

* Averages of three replicate experiments.

Results and Discussion

The pupation percentage (Table I) decreased with increase in concentration of Altozar. The same trend of results was reported by Hongertner and Masner (1973) against the larvae of German cockroach. Skuhravy (1973) reported the mortality (87–98%) of larch cane borer moth with a juvenoid, In the present studies, the adult emergence was 60% in control and 20% in both 0·5% and 0·75% concentrations, while there was no emergence at all at 1·0% of the JHA. The findings are supported by those obtained by Outram (1973) who reported that the emergence was affected at higher doses of a synthetic juvenile hormone (Roeller compound) in *Prodenia litura*. Menn and Beroza (1972) reported that there was no emergence of *Corpocapsa pomponella* when juvenile hormone was applied at 10^{-5} g/cm^2 by contact method.

The adult emergence percentage in respect of pupae was significantly higher in control than in the cases of juvenile hormone treatments.

Outram (1973) observed that there was a low percentage of adult emergence of deformed adults when spruce bud worm pupae were treated with a synthetic juvenile hormone at higher dosages.

In the present study, in general, it was found that the adults emerged from the treated pupae were malformed. This kind of malformations was also reported by Outram (1973) in spruce bud worm, pupae were treated with a juvenile hormone analogue.

The authors are grateful to Prof. G. P. Channa Basavanna for his keen interest and encouragement. The juvenile hormone analogue (Altozar) received from M/s. Zoecon Corporation, Palo Alto, California (U.S.A.), has been gratefully acknowledged.

Department of Entomology, K. C. DEVARAJ URS.
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December 17, 1975.

---

1. Hongertner, M. and Masner, P., *Experiment*, 1973, 29, 1358.
2. Menn, J. J. and Beroza, M., *Insect Juvenile Hormones—Chemistry and Action*. Academic Press, New York and London, 1972 (Edn.), p. 341.
3. Outram, I., *J. Econ. Ent.*, 1973, 66, 1033.
4. Skuhravy, V., *Acta Entomol.*, 1973, 70, 313.

FEEDING EFFECT OF JUVENILE HORMONE ANALOGUE, ZR 619-5 E, ON THE EPILACHNA BEETLE, *EPILACHNA VIGINTIOCTOPUNCTATA* FABRICIUS

JUVENILE hormone analogues (JHA) have been reported to have high biological activity, intermediate specificity, low mammalian toxicity and short environmental persistence. The feeding effect of ZR 619–5 E (Ethyl 11-methoxy-3, 7, 11-trimethyl-2, 4-dodecadiethioate) on the larvae of *Epilachna vigintioctopunctata* has now been investigated.

Materials and Methods

The test insects were from a pure culture maintained in the laboratory on egg plant (brinjal) leaves. Concentrations of 0·25%, 0·50%, 0·75% and 1·00% of ZR 619–5 E were made in 1% acetone and sprayed on individual egg plant leaves. Grubs of third and fourth instars starved for 24 hours were allowed to feed on the treated leaves. Two control regimens, one with 1% acetone and another without any treatment were run alongside. Observations on the feeding behaviour, molting characteristics, etc., of the larvae were made until pupation; fecundity and egg hatchability were also recorded in the case of the adults. The experiment wsa replicated thrice with ten grubs in each The experimental data are presented replication, in Tables I and II.

TABLE I

Effect of ZR 619–5 E on fecundity and per cent hatch of eggs in Epilachna vigintioctopunctata Fb. fed with *treated and untreated egg plant leaves*

Con. ZR 619–5 E%	Av. No. of egg laid	% egg hatch
1·00	182	50
0·75	198	66
0·50	221	67
0·25	260	81
Control (Acetone)	301	95
Blank	312	98

TABLE II

Leaf area scraped by grubs of* Epilachna vigintioctopunctata *Fb.*

Treatment Con. ZR 619–5 E	Area scraped in 24 hrs. in cm²		Area scraped in 48 hrs. in cm²	
	3rd instar	4th instar	3rd instar	4th instar
1·00	0·42	0·3	0·48	0·3
0·75	1·25	0·6	1·6	0·8
0·50	2·4	4·9	3·0	5·6
0·25	3·8	18·5	4·0	20·2
Control (acetone 1%)	40	54	50	68
Control blank	80	86	85	90

* The area treated was converted to 100 cm² and proportional area scraped by 10 grubsin average was calculated.

Results and Discussion

There was a significant reduction in the number of eggs laid and per cent egg hatch of the treated adult beetles as compared to control (Table I). This type of sterility was also reported by Masner (1968) in *Pyrrhocoris* and Riddford (1971) in the silkworm, *Hyalophora cecropia.*

There were no juvenile hormone effect symptoms like, supernumerary molt, prolonged larval instars, failure to pupate, failure to emerge from pupal stage, or formation of abnormal adults, in all the juvenile hormone treatments, except that the first, second and third instar grubs were smaller in size. There was a significant reduction in feeding by grubs on treated leaves as compared to those on untreated leaves (Table II). Bagley and Bauernfein (1972) working with Roeller and Romanuk compounds on citrus snow scale *Unaspis citri* and tobacco worm *Haltothis virescens and* Slama (1973) with some coleoptera and Hymenopteran insects with juvenile hormone analogue, could not obtain the classical symptoms of juvenile hormone.

Conclusion

The juvenile hormone analogue ZR 619–5 E reduced fecundity and per cent hatch of eggs in adults of *Epilachna vigintioctopunctata* Fb. However, in grubs no classical symptoms of juvenile hormone effects could be seen though there was a significant reduction in feeding.

The authors wish to express their deep sense of gratitude to Prof. G. P. Channa Basavanna for his interest and to M/s. Zoecon Corporation, California, U.S.A., for kindly providing the JH analogue, ZR 619–5 E, for this work.

Department of Entomology, K. C. Devaraj Urs.
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December 17, 1975.

---

1. Menn, J. J. and Beroza, N., *Insect Juvenile Hormone : Chemistry and Action,* Academic Press, 1972, pp. 113.
2. Slama, K., *Ann. Rev. Biochem.,* 1971, **40**, 1079.
3. —, Romanuk, M. and Sorm, F., *Insect Hormones and Bioanalogues,* Springer-Verlag, Wien, New York, 1973.
4. Williams, C. M., *Nature,* 1956, **178**, 221.

RING ROT OF BRINJAL CAUSED BY *MYROTHECIUM RORIDUM* TODE EX FRIES FROM BANGALORE

During the months of August and September 1975, it was observed that several fruits of brinjal were affected by ring rot. The extent of damage was 1 to 2%. The disease was noticed at the last stage of the crop and it was more pronounced during last few harvests and was favoured by intermittent rains and sunshine.

The symptoms are evident as small to large conspicuous sharply defined circular to oval flattened lesions. The affected tissues are pale to black. The rot penetrates deep into the fruits. The most characteristic symptom of the disease is the presence of narrow concentric rings of alternating black spore masses and white mycelial mat (Fig. 1).

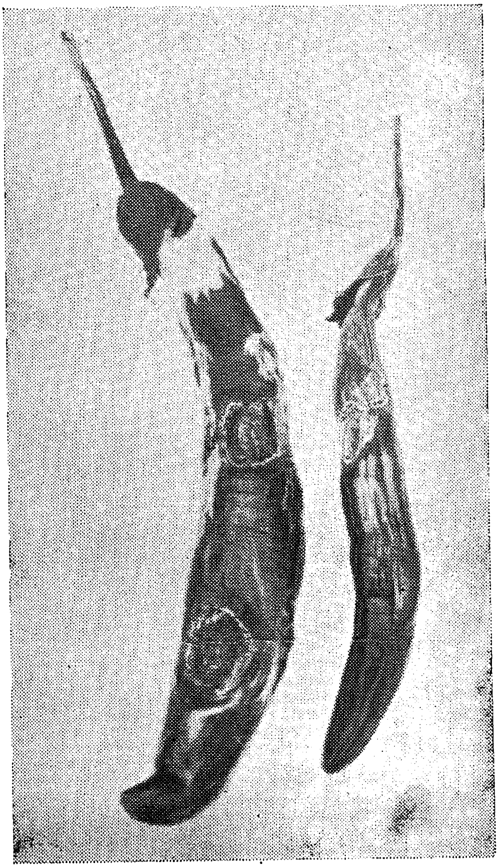

Fig. 1. Symptoms of ring rot on Arka Sheel fruits.

Under artificial inoculation the spores infect both young and mature fruits through wounds, producing characteristic symptom of the disease after 5 days. The following brinjal varieties were found susceptible : Dingra's multiple purple, Pusa kranti, Arka sheel, Arka kusumakar. Ponnappa[1] working with this fungus reported its ability to infect seedlings of 21 economic hosts. This is the first report of *Myrothecium roridum* causing fruit rot of brinjal.

Authors are grateful to the Director, for providing necessary facilities and encouragement.

Indian Institute of Hort. Research,
Bangalore 560 006,
November 21, 1975.

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H. S. SOHI.

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1. Ponnappa, K. M., *Hyacinth Control Jour.*, 1970, 8, 18.

NATIONAL CONFERENCE ON CRYSTALLOGRAPHY (1977)

A National Conference on Crystallography sponsored by the Indian National Committee, for the International Union of Crystallography is proposed to be held at the Department of Physics (Crystallography and Biophysics), University of Madras, from 17th to 21st January, 1977. The year 1977 happens to be the 25th Year of the Department which has been specialising in Crystallography and Biophysics and this programme is a part of the Silver Jubilee activities.

Abstract of papers to be presented at the Conference should reach the Organizing Committee on or before October 15, 1976.

Further particulars may be had from : Prof. R. Srinivasan, Chairman, National Conference on Crystallography, University of Madras, Guindy Campus, Madras 600 025.

INDIAN COUNCIL OF AGRICULTURAL RESEARCH, NEW DELHI-1

Indian Council of Agricultural Research, Krishi Bhavan, New Delhi-1, has evolved a scheme for the appointment of Emeritus Scientists in the field of Agriculture, Animal Husbandry and allied sciences. Nominations are invited for the block year 1977–79. Retired scientists of established reputation are requested to send 15 copies of their biodata and details of the project proposed in the prescribed proforma that can be obtained from Sri. N. Swaminathan, Additional Secretary, ICAR. The last date for receipt of the applications is 30th September 1976.

INDIAN METEOROLOGICAL SOCIETY, NEW DELHI

At a Seminar on Weather Modification held during February, 1976, the current state of knowledge on artificial rain making was reviewed. The Society recognised the need for well planned intensive programme of experiments and made several recommendations in this connection. The details will be published in *Voyu Mandal,* Vol. 6, No. 1.

NEW EVIDENCE OF MAGNETIC POLE VARIATIONS

Scientists, including a team from the Australian National University in Canberra, are uncovering increasing evidence that the earth's magnetic poles have migrated dramatically in the past. The Australian National University's Research School of Earth Sciences' research team has developed techniques which may allow them to reconstruct in detail the variations of the geomagnetic field in Australia over the past 30,000 years. The technique involves taking long, undisturbed oriented cores of sediment from the bottom of lakes and analysing their magnetic records (AIS).

INDIAN SOCIETY OF DEVELOPMENTAL BIOLOGISTS

Indian Society of Developmental Biologists is holding its Scientific Symposium on 2nd January 1977, at Bhubaneswar. Interested Developmental Biologists please contact. : Dr. Suresh C. Goel, Secretary–Treasurer, ISDB, Department of Zoology, Poona University, Poona 411 007.

SHORT SCIENTIFIC NOTES

A New Method of Staining Tissue Sections with Giemsa Using Common Rosin

This method has been found suitable to detect protozoa in the tissue sections, *eg.*, exoerythrocytic schizonts of malaria parasites in the liver, lung or in the kidney. This staining method has been proposed to be named as Giemsa-rosin. Bray and Garnham (1962) used Giemsa-Colophonium method for staining protozoa in tissue sections.

For Giemsa-rosin method the tissue should be fixed in Carnoy's fixative and the sections should not be thicker than 7 μ.

Staining procedure :

The sections are deparaffinished and brought down to tap water and the sections are left for 5 minutes in a weak solution of liquor Ammonia at pH 7·2. The sections are then transferred to the staining solution.

The composition of the staining solution is as follows :

adjusted to pH 7·2 with drops of liquor ammonia

On p. 603, 1st column, after the para "The composition of .. ..as follows :' please *read*—

1. Conc. Giemsa's stain .. 8 ml.
2. Methanol .. 5 ml.
3. Acetone .. 5 ml.
4. Distilled water .. 50 ml.

Also *read* the names of Authors as N. L. Pal and B. Dasgupta.

---

1. Bray, R. S. and Garanham, P. C. C. *Indian J. Malar.*, 1962, 16, 153.

Prodajus ovatus Pillai (Isopoda) Parasitic on a New Host

Bopyrid parasites (family Dajidae) commonly infect Mysids and Euphausiids. In India there is only a single observation by Pillai[1]. Species belonging to the genus *Prodajus* are found parasitic on different *Gastrosaccus* species. Pillai[1] has described *Prodajus gastrosacci* from *Gastrosaccus simulans* and *Prodajus ovatus* from *Gastrosaccus muticus* in the planktons of Tarvancore coast.

Gastrosaccus simulans occurring in the intertidal area at Porto Novo was found infected with Dajid parasites having symmetrical and indistinctly segmented body and reduced pleopods in the female. The highly enlarged anterolateral parts of the peraeon and the drawn out posterolateral parts of lamella present in the parasites collected from *G. simulans* in Porto Novo easily distinguigh it as *Prodajus ovatus* described by Pillai[1]. *G. muticus* has not been so far collected from the waters of Porto Novo. *Prodajus ovatus* occurs on *G. simulans* on the southeast coast of India at Porto Novo while it has been reported from *G. muticus* on the southwest coast of India at Trivandrum.

We are grateful to Prof. R. Natarajan, Director, Centre of Advanced Study in Marine Biology, Porto Novo, for his keen interest and guidance in our work.

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1. Pillai, N. K., *Ann. Mag. nat. Hist.*, 1963, **13** (6), 739.

New Additions to Soil Mycoflora

During the course of investigation of soil mycoflora of a sal forest of Varanasi, two fungal forms, *viz.*, *Beltrania rhombica* (Penzig) Subram[1]. and *Beltraniella-portoricensis* (Stevens) Pirozynski[2] were isolated. These two fungi have not so far been reported from the soil.

Beltramia rhombica (Penzig) Subram.

Colonies on Czapek-yeast extract agar light brown to olive, reverse greenish black. Setae arise from he stroma which are dark, stout and septate 90–135 × 3·5–5 μ in size. Conidiophores arise in groups from the base of the setae, 35–140 × 3–4·5 μ and bear hyaline and oval separating cells, 4–12 × 3–4·5 μ in size. Conidia are olivaceous green, biconic with a hyaline band on their broadest part, 21–25·5 × 7–10·5 μ; their proximal end is V-shaped and distal end furnished with a hyaline seta 7–10 μ in length.

Beltraniella portoricensis (Stevens) Pirozynski.

Colonies on Czapek-yeast extract agar pale olivaceous grey to dark brown, reverse black. Setae are olive brown septate branched and 160–400 × 3·25–3·5 μ in size. Conidiophores arise as lateral branches from the setae just below the septum, are thin walled bearing separating cells 10–18 × 3·2–3·5 μ Conidia are acrogenous smooth walled, sub-hyaline, 17·5–21·0 × 5·2–7·5 μ, having a hyaline band at their broadest region.

This specimen has been deposited in C.M.I., herb. No. IMI 175124.

We are thankful to Dr. R. Y. Roy for suggestions and one of us (ANS) acknowledges gratefully the financial assistance from C.S.I.R., New Delhi.

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January 20, 1976.

---

1. Pirozynski, K. A. and Patil, S. D., *Cand. J. Botany*, 1970, 48 (3), 567.
2. Subramanian, C. V., *Hyphomycetes*, ICAR, New Delhi, 1971, p. 888.

Some New Leaf Spot Diseases

During the year 1975–76 the authors have observed three leaf spot diseases, caused by pathogenic fungi in the local botanical garden. The pathogens causing diseases were isolated separately by the usual methods. The cultures of different fungi were identified and have been deposited in the Botany Department, Bhagalpur University. The morphological characters of the organisms were similar to those of type species. The symptoms of the diseases are given below:

Alternaria alternata (Fr.) Keissler produced dark brown necrotic spots on the leaves of *Melia azadirachta* Linn. The lesions were irregular in shape and 2–5 mm in diameter. Concentric rings of conidial mass appeared on the older spots.

A leaf spot of *Mimusops elengi* Linn. incited by *Epicocum purpurescens* Ehrenb. was observed during January and February 1976. The spots were marginal, light reddish brown and 10·15 mm in diam.

The severe anthracnose disease on the leaves of *Cinnamomum tamala* F. Nees. caused by *Colletotrichum gloeosporioides* Penz. was noticed during October 1975. The lesions usually started from tips and proceeded downward marginally. The diseased area was drab brown and covered the entire surface of the leaf.

The pathogenicity tests on their respective hosts were found to be satisfactory. The above-mentioned diseases are new records from India[1–4].

Sincere thanks are due to Professor K. S. Bilgrami, Head, Department of Botany, Bhagalpur University, for providing laboratory facilities and guidance.

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March 12, 1976.

---

1. Butler, E. J. and Bisby, G. R., *The Fungi of India*, Issued by I.C.A.R., New Delhi, 1931–32.
2. Sarbhoy, A. K., Lal, G. and Varshney, J. L., *Ibid.*, Published by Navyug Traders New Delhi. 1967–71, pp. 150,
3. Tailak, S. T. and Rao, R., *Fungi of India Supplement* 2, 1962–67.
4. Vasudeva, R. S., *Fungi of India Supplement* 1, Issued by I.C.A.R., New Delhi, 1953–57.

Trace Element Nutrition of Mushroom *Pleurotus sajor-caju* (Fr.) Singer

The trace element requirements of cultivated mushroom (*Agaricus bisporus*) have already been studied[1,4]. The information regarding the trace element nutrition of other edible fungi is rather lacking. The present information deals with the trace element nutrition of *Pleurotus sajor-caju* which is a well-known edible mushroom and is condidered a delicacy.

Five trace elements (ferrous sulphate 2 ppm, zinc sulphate 1 ppm, manganese sulphate 0·5 ppm, molybdenum as ammonium molybdate 3 ppm and boron as potassium bromide 5 ppm) were tried to see their effect on the growth of the mushroom. Trace elements present in various analytical grade chemicals as impurities were removed by the Steinberg's method[3]. The pH of the basal medium (Czepek Dox) was adjusted to 5·6 and 5 ml of mycelial suspension was used for inoculation. The inoculated flasks were incubated at 25° C and mycelium harvested after 12 days.

Addition of ferrous sulphate individually resulted in the maximum growth and this was followed by zinc sulphate. When ferrous sulphate and zinc sulphate were added in different concentrations (0·1, 1, 5, 10 ppm) individually as well as collectively, maximum dry mycelial weight was harvested when mixture of ferrous sulphate and zinc sulphate was used in various concentrations. Addition of ferrous sulphate singly yilded maximum growth at a concentration of 10 ppm/1. But zinc sulphate upto a concentration of 1 ppm/1. increased growth and at higher (5–10 ppm/l.) concentrations there was no appreciable increase in growth. The present observations are in good agreement with the findings of Humfeld and Sugihara[2] in the case of *Agaricus campestris*.

Department of Botany and C. L. Jandaik.
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1. Bhous, G., *Mushroom Sci.*, 1959, **4**, 86.
2. Humfeld, H. and Sugihara, T. F., *Mycologia*, 1952, **44**, 605.
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REVIEWS AND NOTICES OF BOOKS

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It is very well recognised that meteorological factors ay a key role in the distribution and build up of sects. Understanding these factors help in fore-sting outbreak of pests and thereby managing them. he Colorado potato beetle originating somewhere the central part of the USA and which had its begin-ng in 1820s is now found in almost all temperate gions of America and Europe. This bulletin has x chapters, and includes three appendices also.

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G. P. CHANNABASAVANNA.

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The articles deal with the methods developed during the last few years for the estimation of heavy metal pollutants. The heavy metals covered are mercury, lead, cadmium, copper, zinc, antimony, cobalt. nickel, silver, iron, manganese, vanadium etc. Some non-metals like, phosphorous and sulphur, are also covered. Major portion of the book is devoted to the analytical aspects of mercury. It highlights the sources of mercury pollution, both from natural sources and due to human activity, and the consequent health and ecological problems. The usefulness of mercury and its safety limits are assessed making use of relevant facts. A critical review of various methods suitable for the determination of mercury in minute amounts in food, environment and animal tissues is given and this is very useful for both scientists as well as administrators who may have something to do with mercury estimation and mercury pollution.

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R. NARAYANA AND G. NAGENDRAPPA.

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Professional designers and other users of the Delphi method will find in this book the much needed background information on the technique. Hopefully the volume will help them to effectively use the Delphi method.

Mrs. K. Rohatgi.
P. K. Rohatgi.

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Dr. Richard Jago of Monash University, Melbourne, cooled a piece of iron meteorite in liquid nitrogen and noticed certain changes in the meteoric metal. He concludes that these meteors were never subjected to low temperatures (about − 269°) prevailing outside the solar system and hence the origin of the meteors must be within the solar system where the minimum temperature is round about *minus* 100° C (AIS).

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and Printed at The Bangalore Press, Bangalore-560018.

SPECTRAL SHAPES OF $1^- \rightarrow 2^+$ AND $1^- \rightarrow 0^+$ BETA TRANSITIONS OF ^{194}Ir

K. VENKATA RAMANAIAH, G. KUSA RAJU AND K. VENKATA REDDY
Laboratories for Nuclear Research, Andhra University, Waltair, India

ABSTRACT

The spectral shapes of the two intense beta groups of ^{194}Ir are studied with a thoroughly tested Siegbahn-Slatis beta ray spectrometer of the intermediate image type. The $1^- \rightarrow 2^+$ beta transition with an end point energy of 1921 ± 2 KeV and the $1^- \rightarrow 0^+$ beta transition with its end-point energy 2251 ± 2 KeV are surprisingly statistical in spite of their high log ft. values. The 1921 KeV beta transition cannot be analysed under modified B_{ij} approximation and the shapes of the two beta transitions are of identical nature. The beta transitions are in accordance with ξ-approximation supporting the earlier results.

INTRODUCTION

^{194}Ir decays to ^{194}pt with a half-life of 17 hours. A spin sequence of $1^- \rightarrow 2^+$ and $1^- \rightarrow 0^+$ was established[1] for the first inner and ground state beta transitions.

The 1920 KeV beta component from ^{194}Ir feeding the first excited state of ^{194}pt with 329 KeV energy has an intensity of 5·1% and log ft. value 9·2. The ξ-value ($\alpha Z/2R$) for this is 10·4 and is much greater than $(W_0 - 1) = 3{\cdot}758$, where, W_0 is the end-point energy expressed in $m_0 c^2$ units. The criterion for the applicability of ξ-approximation, $\xi \gg W_0 - 1$ is well fulfilled in this case. The directional correlation of the 1920 KeV beta-384 KeV gamma cascade shows from the results of Deutsch *et al.*[2] small beta-gamma anisotropy. Recently, Priyadarsini[3] reported zero anisotropy for this cascade. There has been only one measurement on the shape of the $1^- \rightarrow 2^+$ beta transition by Deutsch *et al.*[2] and the shape of the $1^- \rightarrow 0^+$ beta transition with a log ft. value of 8·2 has not yet been measured. The large log ft. value and a possible deviation from ξ-approximation indicate the possibility of the application of modified B_{ij} approximation for the $1^- \rightarrow 0^+$ beta transition. In order to check the applicability of the modified B_{ij}-approximation, conclusive evidence as to whether the spectrum shape factor is energy-independent is necessary. For this purpose a shape analysis of the two beta transitions is carried out.

EXPERIMENTAL DETAILS AND RESULTS

^{194}Ir sources are supplied by Bhabha Atomic Research Centre, Bombay, in three consignments, each time irradiating samples of iridium at CIRCUS reactor. There, we found strong contamination of ^{192}Ir. Since the maximum beta energy in the decay of ^{192}Ir is only 662 KeV, there will be no interference with the present studies. Sources are prepared on aluminised mylar foils of thickness ~ 180 μg/cm^2. Insulin is used for uniform spreading of the source.

The Siegbahn-Slatis beta ray spectrometer, presently used, has been described by Nagarajan *et al.*[4,5] with an emphasis on its adaptability for the studies of small-order deviations in shape factors of ^{90}Y and ^{198}Au. The present mode of analysis of beta spectra has also been given by Nagarajan *et al.*[5].

The beta spectrum is scanned in steps of ~ 15 KeV from 1,500 KeV to 2,300 KeV only in the singles mode of operation of the spectrometer. Background at each measurement point is taken, taking the advantage of the special provision of the present spectrometer for closing the central baffle of the spectrometer thus shielding the detector from the source without disturbing the vacuum. A computer program 'FERMKURI' takes care of the corrections due to half-life, and other instrumental effects like back-scattering correction, correction due to finite resolution, etc. 'FERMKURI' also incorporates the electron radial wave functions of Bhalla and Rose[6] and the screening corrections due to Buhring[7].

A current setting is adjusted corresponding to 1900 keV and the count rate is followed for five half-lives. A least squares fit of the data yields 17 ± 0·04 hrs. for the half-life of ^{194}Ir.

Analysis of $1^- \rightarrow 0^+$ *Beta Transition*

There is only a region of 350 KeV for the analysis of $1^- \rightarrow 0^+$ outermost beta transition. The exact end-point energy for this beta group from the physical behaviour of the shape factor plot [$N/p^2F(Z, W)(W_0 - W)^2$ *vs* energy (where W_0 is the end-point energy) near W_0] the end-point energy was found to be 2251 ± 2 KeV. This exact end-point energy is used in a further analysis through a computer program 'SHAPFIT' and the experimental points are found to fit very well with a shape factor of the form $K(1 + aW)$ with $a = 0{\cdot}03 \pm 0{\cdot}06$. Three such runs with sources of different diameter are taken. The results of the three runs are given in Table I. The shape factor plot due to one of the runs is given in Fig. 1.

Analysis of $1^- \rightarrow 2^+$ *Beta Transition*

The outermost beta transition corrected for its shape is subtracted from the gross-spectrum of ^{194}Ir with an end-point energy of 2251 ± 2 KeV using the relation $Y_2 = \sqrt{Y^2 - Y_1^2 C_1}$ where Y_2 and Y_1 are the ordinates of the outer and inner beta groups in the 'FERMI-KURIE' plots. The resulting spectrum from 600 keV and 1920 KeV is subjected shape analysis[4]. The branches from the setae just below the septum, are thin walled bearing separating cells 10–18 × 3·2–3·5 μ Conidia are acrogenous smooth walled, sub-hyaline, 17·5–21·0 × 5·2–7·5 μ, having a hyaline band at their broadest region.

This specimen has been deposited in C.M.I., herb. No. IMI 175124.

We are thankful to Dr. R. Y. Roy for suggestions and one of us (ANS) acknowledges gratefully the financial assistance from C.S.I.R., New Delhi.

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January 20, 1976.

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1. Pirozynski, K. A. and Patil, S. D., *Cand. J. Botany*, 1970, 48 (3), 567.
2. Subramanian, C. V., *Hyphomycetes*, ICAR, New Delhi, 1971, p. 888.

Some New Leaf Spot Diseases

During the year 1975–76 the authors have observed three leaf spot diseases, caused by pathogenic fungi in the local botanical garden. The pathogens causing diseases were isolated separately by the usual methods. The cultures of different fungi were identified and have been deposited in the Botany Department, Bhagalpur University. The morphological characters of the organisms were similar to those of type species. The symptoms of the diseases are given below:

Alternaria alternata (Fr.) Keissler produced dark brown necrotic spots on the leaves of *Melia azadirachta* Linn. The lesions were irregular in shape and 2–5 mm in diameter. Concentric rings of conidial mass appeared on the older spots.

A leaf spot of *Mimusops elengi* Linn. incited by *Epicocum purpurescens* Ehrenb. was observed during January and February 1976. The spots were marginal, light reddish brown and 10·15 mm in diam.

The severe anthracnose disease on the leaves of *Cinnamomum tamala* F. Nees. caused by *Colletotrichum gloeosporioides* Penz. was noticed during October 1975. The lesions usually started from tips and proceeded downward marginally. The diseased area was drab brown and covered the entire surface of the leaf.

The pathogenicity tests on their respective hosts were found to be satisfactory. The above-mentioned diseases are new records from India[1-4].

Sincere thanks are due to Professor K. S. Bilgrami, Head, Department of Botany, Bhagalpur University, for providing laboratory facilities and guidance.

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Bhagalpur–812007. M. M. PRASAD.
March 12, 1976.

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1. Butler, E. J. and Bisby, G. R., *The Fungi of India*, Issued by I.C.A.R., New Delhi, 1931–32.
2. Sarbhoy, A. K., Lal, G. and Varshney, J. L., *Ibid.*, Published by Navyug Traders New Delhi. 1967–71, pp. 150,
3. Tailak, S. T. and Rao, R., *Fungi of India Supplement* 2, 1962–67.
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Five trace elements (ferrous sulphate 2 ppm, zinc sulphate 1 ppm, manganese sulphate 0·5 ppm, molybdenum as ammonium molybdate 3 ppm and boron as potassium bromide 5 ppm) were tried to see their effect on the growth of the mushroom. Trace elements present in various analytical grade chemicals as impurities were removed by the Steinberg's method[3]. The pH of the basal medium (Czepek Dox) was adjusted to 5·6 and 5 ml of mycelial suspension was used for inoculation. The inoculated flasks were incubated at 25° C and mycelium harvested after 12 days.

Addition of ferrous sulphate individually resulted in the maximum growth and this was followed by zinc sulphate. When ferrous sulphate and zinc sulphate were added in different concentrations (0·1, 1, 5, 10 ppm) individually as well as collectively, maximum dry mycelial weight was harvested when mixture of ferrous sulphate and zinc sulphate was used in various concentrations. Addition of ferrous sulphate singly yilded maximum growth at a concentration of 10 ppm/1. But zinc sulphate upto a concentration of 1 ppm/1. increased growth and at higher (5–10 ppm/l.) concentrations there was no appreciable increase in growth. The present observations are in good agreement with the findings of Humfeld and Sugihara[2] in the case of *Agaricus campestris*.

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In a brief introduction in chapter 1 an account of how the Colorado potato beetle made its beginning and spread to most parts of temperate America and Europe is presented. The life-cycle of the beetle, which largely depends on temperature, is briefly reviewed in chapter 2. In the warmer areas of its distribution the insect can pass through four generations, whereas in the colder parts one full and one partial generations are possible, and in the extreme cold areas only a partial generation and this means the beetle cannot establish in the latter regions.

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K. VENKATA RAMANAIAH, G. KUSA RAJU AND K. VENKATA REDDY
Laboratories for Nuclear Research, Andhra University, Waltair, India

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^{194}Ir sources are supplied by Bhabha Atomic Research Centre, Bombay, in three consignments, each time irradiating samples of iridium at CIRCUS reactor. There, we found strong contamination of ^{192}Ir. Since the maximum beta energy in the decay of ^{192}Ir is only 662 KeV, there will be no interference with the present studies. Sources are prepared on aluminised mylar foils of thickness ~ 180 μg/cm^2. Insulin is used for uniform spreading of the source.

The Siegbahn-Slatis beta ray spectrometer, presently used, has been described by Nagarajan *et al.*[4,5] with an emphasis on its adaptability for the studies of small-order deviations in shape factors of ^{90}Y and ^{198}Au. The present mode of analysis of beta spectra has also been given by Nagarajan *et al.*[5].

The beta spectrum is scanned in steps of ~ 15 KeV from 1,500 KeV to 2,300 KeV only in the singles mode of operation of the spectrometer. Background at each measurement point is taken, taking the advantage of the special provision of the present spectrometer for closing the central baffle of the spectrometer thus shielding the detector from the source without disturbing the vacuum. A computer program 'FERMKURI' takes care of the corrections due to half-life, and other instrumental effects like back-scattering correction, correction due to finite resolution, etc. 'FERMKURI' also incorporates the electron radial wave functions of Bhalla and Rose[6] and the screening corrections due to Buhring[7].

A current setting is adjusted corresponding to 1900 keV and the count rate is followed for five half-lives. A least squares fit of the data yields 17 ± 0·04 hrs. for the half-life of ^{194}Ir.

Analysis of $1^- \rightarrow 0^+$ *Beta Transition*

There is only a region of 350 KeV for the analysis of $1^- \rightarrow 0^+$ outermost beta transition. The exact end-point energy for this beta group from the physical behaviour of the shape factor plot [N/p^2F (Z, W) $(W_0 - W)^2$ *vs* energy (where W_0 is the end-point energy) near W_0] the end-point energy was found to be 2251 ± 2 KeV. This exact end-point energy is used in a further analysis through a computer program 'SHAPFIT' and the experimental points are found to fit very well with a shape factor of the form K $(1 + aW)$ with $a = 0 \cdot 03 \pm 0 \cdot 06$. Three such runs with sources of different diameter are taken. The results of the three runs are given in Table I. The shape factor plot due to one of the runs is given in Fig. 1.

Analysis of $1^- \rightarrow 2^+$ *Beta Transition*

The outermost beta transition corrected for its shape is subtracted from the gross-spectrum of ^{194}Ir with an end-point energy of 2251 ± 2 KeV using the relation $Y_2 = \sqrt{Y^2 - Y_1{}^2 C_1}$ where Y_2 and Y_1 are the ordinates of the outer and inner beta groups in the 'FERMI-KURIE' plots. The resulting spectrum from 600 keV and 1920 KeV is subjected shape analysis[4]. The

TABLE I

Shape factor results $1^- \rightarrow 0^+$ *beta transition of* ^{194}Ir

Run No.	End-point energy (KeV)	C (W)/K
1	2251 ± 2	1 + (0·026 ± 0·06) W
2	2250 ± 2	1 + (0·032 ± 0·06)W
3	2251 ± 2	1 + (0·025 ± 0·06)W

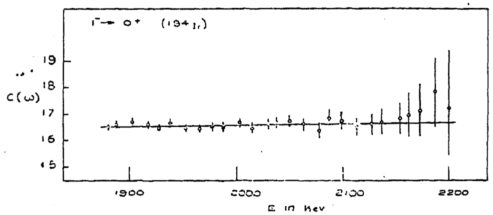

FIG. 1. The shape factor plot of the 2250 KeV beta transition of ^{194}Ir. Solid line is the least-square fit with C (W) = K (1 + *a*W) where *a* = + 0·0266 ± 0·058.

'BETASHAP' has given an exact end-point energy of 1920 ± 2 KeV. The experimental points are once again found to be in accordance with statistical shape in this case also, fitting into a shape of the form K (1 + *a*W) with *a* = 0·025 ± 0·04 with E_0 = 1920 ± 2 KeV. The shape factor plot for one of the runs is shown in Fig. 2. The error bars also include a factor of 1·25 due to the subtraction of the outer beta group. The results for the three runs are given in Table II.

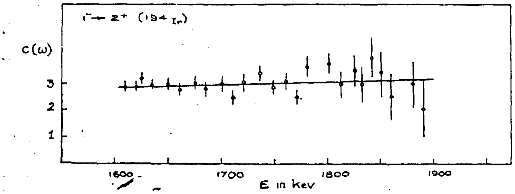

FIG. 2. Shape of the 1920 KeV beta transition of ^{194}Ir. Solid line is the least-squares fit with C(W) = C (1 + *a*W) with *a* = 0.031 ± 0·038. The error bars include a factor of 1·25 due to the subtraction of outer beta group.

TABLE II

Shape factor results of $1^- \rightarrow 2^+$ *beta transition of* ^{194}Ir

Run No.	End-point energy (KeV)	C (W)/K
1	1920 ± 2	1 + (0·031 ± 0·04) W
2	1920 ± 2	1 + (0·022 ± 0·04) W
3	1920 ± 2	1 + (0·023 ± 0·04) W

DISCUSSION

The log ft. values of the main beta transitions, of 8·2 and 9·2, suggest the assignment of negative parity to the ground state of ^{194}Ir and hence the first forbidden nature of the two beta transitions. The shape of the 1920 KeV beta transition is statistical within the experimental errors. In an analysis employing modified B_{ij}-approximation[9,10], the shape factor is expected to have a unique component. Hence the modified B_{ij}-approximation cannot be applied in this case.

From the present measurements within the experimental accuracy, the shape factors of both the $1^- \rightarrow 2^+$ and $1^- \rightarrow 0^+$ beta transitions are fitting into the same shape factor form C (W) = K (1 + *a*W) and at the same time exhibiting statistical nature. Since the B_{ij} matrix element cannot contribute to $1^- \rightarrow 0^+$ beta transition and since the shapes are identical, it can be said that the shape of the $1^- \rightarrow 2^+$ beta transition is not determined by B_{ij} matrix element. This is also supported by the angular distribution measurements of Brewer *et al.*[8], wherein the unique matrix element B_{ij} in the $1^- \rightarrow 2^+$ beta transition to the 3·28 KeV level of ^{194}pt is small or zero.

In fact, the equality of the shape factors of the inner and the outer beta branches is not unexpected in the framework of the rotational model assuming the same matrix elements to contribute for beta transitions to various members of a rotational band. In order to arrive at a better physical understanding of the present measurements, an analysis of the data in terms of the matrix elements is required.

From the present measurements of the beta spectral shapes and from the angular correlation measurements of Priyadarsini[3], it may be concluded that 1920 KeV beta transition of ^{194}Ir follows the ξ-approximation[11]. It is normally difficult to obtain nuclear matrix elements of the beta transition following ξ-approximation unless the number of available experimental observables (data on different types of polarisation) is large.

ACKNOWLEDGEMENTS

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1. *Nuclear Data Tables*, 1972, 73, 95.
2. Duetsch, J. P., Grenacs, L. and Lehmann, J., *Kolloguim Uber beta Zerfall and Schwache Wechselwirkung*, Heidelburg, 1965, p. 257.
3. Priyadarsini, D. K. *et al.*, *Curr. Sci.*, 1975, **44**, 22, 793.
4. Nagarajan, T., Ravindranath, M. and Venkata Reddy, K., *Nucl. Instr. and Methods*, 1969, **67**, 77.
5. — and Venkata Reddy, K., *Ibid.*, 1970, **80**, 217.
6. Bhalla, C. P. and Rose, M. E., *ORNL Report*, 1962, p. 3207.
7. Buhring, W., *Nucl. Phys.*, 1965, **61**, 110.
8. Brewer, W. D. and Shirly, D. A., *Ibid.*, 1970, **A 149**, 392.
9. Morita, R. and Morita, M., *Phys. Rev.*, 1958, **109**, 2048.
10. Matumato, Z., Morita, M. and Yamada, M., *Bull. Motayasi, Inst. Phys. Res.*, 1955, **5**, 210.
11. Kotani, T. and Ross, M., *Phys. Rev.*, 1959, **113**, 623.

FIRST RECORD OF FILAMENTOUS ALGAL REMAINS FROM THE LATE PRECAMBRIAN ROCKS OF RANDOM ISLAND (TRINITY BAY), EASTERN NEWFOUNDLAND, CANADA

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ABSTRACT

The late Precambrian shales of the Trinity Bay area (Random Island) of Eastern Newfoundland, Canada, yielded a rich concentration of filamentous algal remains (*Siphonophycus kestron*, *Siphonophycus* sp. A, *Gunflintia* sp. A, *Heliconema* sp. A) of living cyanophycean (nostocalean) affinities. The general morphological characters of these four species of ancient algae are described. Their discovery in these rocks of the area is the first find of these groups of microfossils known to-date. The presence of these microorganisms in the rocks assists in the reconstruction of paleoenvironments, dating of Precambrian sediments and in delineation of boundary between the Precambrian and Cambrian sequences of the area.

INTRODUCTION

THE occurrence of fossil filamentous algal remains from the Precambrian sedimentary rocks of Newfoundland, Canada, has not been reported so far. In Canada, however, Barghoorn and Tyler (1965 *a*, *b*) have recorded microorganisms, including algal filaments, from the Gunflint chert (Gunflint Iron Formation, Animikie Series), of Middle Precambrian age. Similarly, Hofmann and Jackson (1969) also observed microfossils (chains and clumps of bacteria, filamentous and spheroidal structures of probable algal or fungal affinities) from the dark gray chert (Belcher Group) of Hudson Bay, Ontario.

It may be interesting to note that the Australian Precambrian sediments have revealed an amazingly rich and varied assemblage of microorganisms, including algal filaments (Barghoorn and Schopf, 1965; Schopf and Barghoorn, 1968; Schopf and Barghoorn, 1969; Schopf, 1970; Schopf and Blacic, 1971; Schopf, 1972), which are universally well known at present. The present paper reports the first discovery of filamentous algal remains from the Late Precambrian rocks exposed on Random Island in the Trinity Bay area of Eastern Newfoundland, Canada.

In Eastern Newfoundland (Canada), a fairly thick cross-section of the Precambrian to Lower Paleozoic sedimentary rocks (of the Appalachian Mountain system) occurs in the Trinity Bay area of Atlantic ocean. The exposures of the algal filament bearing shale beds are confined between the coordinates 48° 06′ 35″ N to 48° 06′ 45″ N and coordinates 53° 00′ 53′ 30″ W to 53° 00′ 53′ 50″ W of Random Island in Trinity Bay (Table I).

MORPHOLOGICAL DESCRIPTION

The algal filaments recovered through maceration from the medium gray shales are well preserved and are medium to dark brown in colour. The general morphological characters of the four species of ancient algae discovered are given below.

Systematic Descriptions and Biological Relations of algae:

Phylum: CYANOPHYTA, Class: CYANOPHYCEAE, Order: NOSTOCALES, Family: OSCILLATORIACEAE (S. F. Gray) Dumortier ex Krichner, 1988,

Genus: *Siphonophycus* Schopf, 1968,

Siphonophycus kestron Schopf, 1968,

Figs. 10 and 11

Thallus broad, tubular, moderate size, unbranched, non-septate, straight to slightly bent in nature, finely rugose, 42 to 45 μ long (in partly broken specimens), apices partly capitate (apparent in a specimen), in places more or less constricted adjacent to expanded parts, broadly conical with a bluntly pointed terminus; thallus 6 to 7 μ wide, finely microgranulose throughout, ornamented and ringed by finely punctate surficial ridges (*see* arrow in Figs. 10, 11) regularly spaced about 2/3 μ apart.

Siphonophycus sp. A

Figs. 1–9

Thallus broad, tubular, non-septate, unbranched, straight to slightly bent, 30 to about 95 μ long (in partly broken specimens), slight constrictions rarely seen in places throughout; however, constriction (of 2·50 to 4·50 μ across) distinct at one end of thallus (*see* arrow in Figs. 1, 4, 5), other end tapering to a sharp point with a slightly curved hook; thallus 6 to 9·50 μ wide, commonly 6 μ wide, finely microgranulose (with diameter of granules 0·50 μ) throughout.

Genus *Heliconema* Schopf, 1968

Heliconema sp. A

Figs. 12–16

Thallus tubular, flattened just like a ribbon, non-septate, unbranched, loosely coiled at an angle of approx. 45° into a helical fashion, spirals (*see* arrow in Figs. 15, 16) commonly about 6 μ broad and set apart from the adjacent ones; apices not attenuated and flat-rounded; surface texture finely microgranulose (granules up to 0·50 μ diameter); thallus 5–10 μ wide, commonly about 6 μ wide, up to 85 μ long; coiled thallus nearly straight (Fig. 13).

Genus *Gunflintia* Barghoorn, 1965

Gunflintia sp. A

Fig. 17

Thallus broad, tubular, septate (*see* arrow in Fig. 17), unbranched, straight to curved, about 114 μ long (in a partly broken specimen), more or less slightly constricted adjacent to slightly expanded parts of thallus;

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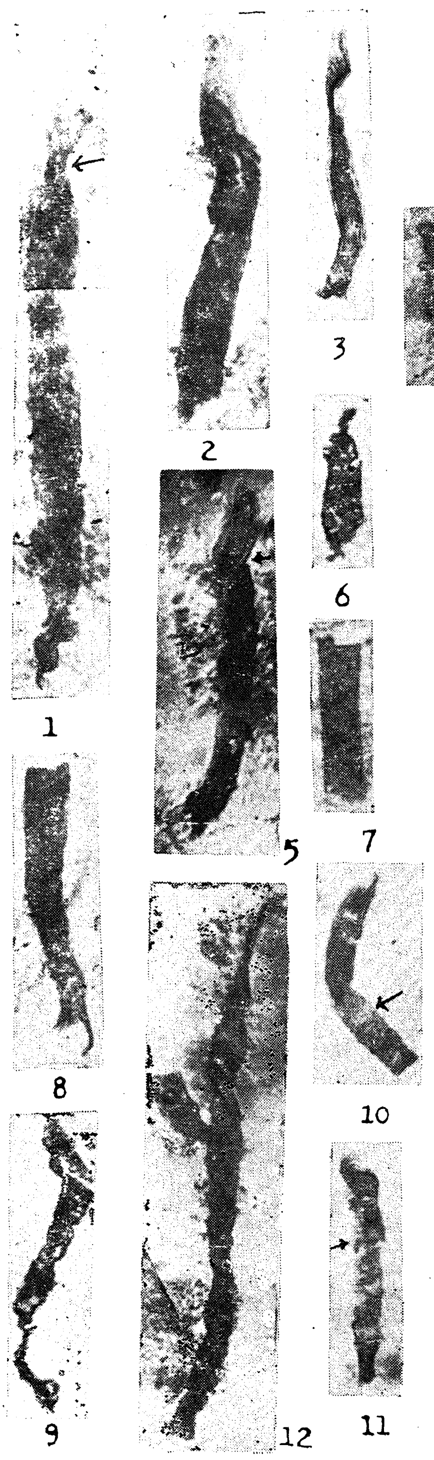

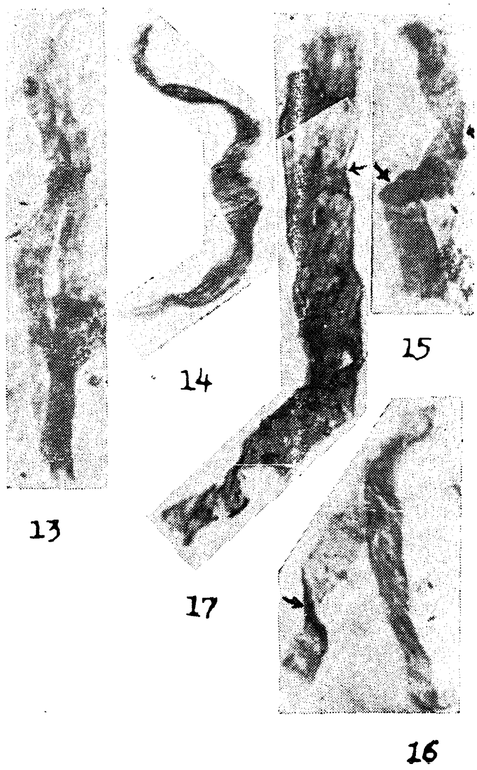

FIGS. 1–17. Photomicrographs of filamentous algal remains from late Precambrian shales of Random Island in Trinity Bay of Eastern Newfoundland, Canada. Figs. 1–9. *Siphonophycus* sp. A; Figs. 10, 11. *S. kestron*; Figs. 12–16. *Heliconema* sp. A; Fig. 17. *Gunflintia* sp. A. All Figs., × 2,000 approx.

thallus about $12\,\mu$ wide, finely and densely microgranulose throughout (diameter of granules less than $0 \cdot 50\,\mu$), septae distinct and very thin (about $0 \cdot 50\,\mu$ thickness), apart from adjacent ones by about $15\,\mu$ distance; individual filaments of commonly uniform diameter throughout but in places with slightly constricted parts.

PALAEOENVIRONMENT

The occurrence of these filamentous, benthic algal remains discloses some facts about their close comparison with the living nostocalean blue-green algae which may be referred as lower thallophytes. They are of cyanophycean (nostocalean) affinities.

The development of benthic, filamentous blue-green algae requires the process of photosynthesis. Their rich concentration in the sediment is suggestive of existence of the extensive shallow ocean that provided a highly favourable environment (littoral photic zone) for algal growth. A similar view is also shared by Schopf and Blacic (1971) for the development of blue-green algae in the late Precambrian sediments (Bitter Spring Formation) of North-Central Amadeus Basin of Australia. Furthermore, this benthic assemblage is associated with the rare microorganisms (*Leiosphaeridia* sp. 1 types 1, 2 and *Leiovalia ovalis*, Table I) to planktonic habitat of shallow marine environment.

AGE ASSIGNMENT AND CORRELATION

There is a high concentration and dominance, in quantitative composition, of blue-green algal filaments (Figs. 1–17) with some fungal remains in the medium gray shales (Unit No. 33, Table I) of the Trinity Bay area. Schopf (1970, 1972) and Schopf and Blacic (1971) also suggest that the blue-green algae had reached their zenith during the late Precambrian time. Their view on the occurrence of ancient blue-green algae also confirms the authors proposal that the medium gray shales are undoubtedly of late Precambrian in age. In addition, these algal forms (*Gunflintia* sp. A, *Heliconema* sp. A, *Siphonophycus* sp. A, *S. kestron*) have not been recorded from the Cambrian sequences of any part of the world to-date. They are the representatives of the late Precambrian sequences (Schopf 1970, 1972; Schopf and Blacic, 1971).

The profuse concentration and confined occurrence of blue-green algae in the uppermost late Precambrian sequence (Unit No. 33, Table I) of Eastern Newfoundland (Trinity Bay area) is of great significance, suggesting that they can profitably be used as " biostratigraphic markers " for the biostratigraphic correlation of the Precambrian sequences (upper part). Furthermore, their presence in the sediments assists to delineate the boundary between the Precambrian and Cambrian sequences (Table I).

ACKNOWLEDGEMENT

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TABLE I

General stratigraphic succession of Precambrian (uppermost part) and Early Cambrian (lowermost part) sequence at northern Northwest Arm section of Random Island in Trinity Bay, Eastern Newfoundland, Canada, with algal remains. Formational nomenclature after Jenness[5] (colour names given to rocks are taken from *Rock-Colour Chart* distributed by the Geological Society of America, 1963).

Lithological characters and microflora
Approximate thickness of units studied (in feet)

ADEYTON GROUP
Chamberlain's Brook Formation:
Nautiyal's Rock Unit No. 31,

Shale, greenish gray, non-calcareous, compact and hard; acritarchs, *Leiosphaeridia* sp. l, type 1, *Leiosphaeridia* sp. l., type 2, *L.* cf. *granulata*, and *Leiofusa filifera* 20

Unit No. 32,

Shale and Quartzite; Shale, medium gray, non-calcareous, partly papery, quartzitic, with thin bands of medium grained and greenish gray quartzite; acritarchs, *Archaeo hystrichosphaeridium* sp. 2, *Baltisphaeridium* sp. 1, *Leiosphaeridia* sp.l., type 1, *Leiosphaeridia* sp. 1, type 2, *Leiofusa filifera*, *Multiplicisphaeridium lancarae*, *M. martae*, *Nucellosphaeridium* sp. 2, and *Orygmatosphaeridium* sp. 1.....5

Units 31 and 32, Early Cambrian age (after Jennes[5] and also by Nautiyal method of this paper).

Random Formation:
Unit No. 33,

Shale, medium gray, non-calcareous, compact and hard, occasionally with some black carbonaceous inclusions; acritarchs, *Leiosphaeridia* sp. 1, type 1, *Leiosphaeridia* sp. 1, type 2, *Leiovalia ovalis*; filamentous algal remains, *Gunflintia* sp. A, *Heliconema* sp. A, *Siphonophycus* sp. A and *S. kestron*. Late Precambrian age (after Nautiyal method of this paper). 5

Unit No. 34,

Shale, medium gray, non-calcareous, compact and hard, a fault (?) contact seems to be present between the Units 33 and 34; acritarch, *Nucellosphaeridium* sp. 1. 12

Unit No. 35,

Chert, light olive gray, non-calcareous, with sporadic distribution of fine grains of pyrite; phytoplankton, *Huronispora* sp. 1, *H. psilata*; acritarch, *Nucellosphaeridium* sp. 1. 3

Units 34 and 35, Late Precambrian age.

Lower part of Precambrian sequence not incorporated.

1. Barghoorn, E. S. and Schopf, J. W., *Science*, 1965, **150**, 3694, 337, 339.
2. — and Tyler S. A., *Current Aspects of Exbiology*, Pergamon Press, Oxford, 1965 *a*, **93**, 118.
3. — and —, *Science*, 1965 *b*, **147**, 3658, 563, 577.
4. Hofmann, H. J. and Jackson, G. D., *Can. Jourl. E. Sciences*, 1969, **6** (5), 1137, 1144.
5. Jenness, S. E., *Geol. Surv. Canada*, 1963, Mem. 327, 30, 31, 59.
6. Schopf, J. W. and Barghoorn, E. S., *Jourl. Paleont.*, 1969, **43**, 1, 111, 118.
7. —, *Biol. Rev.*, 1970, **45**, 319, 352.
8. — and Blacic, J. M., *Jourl. Paleont.*, 1971, **45** (6), 925, 960.
9. —, *Proc. XXIV Int. Geol. Congress*, 1972, **1**, 68, 77.

OCTAHEDRAL (sp^3d^2) NICKEL (II) COMPLEXES OF AROYL HYDRAZONES

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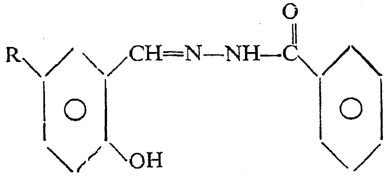
ABSTRACT

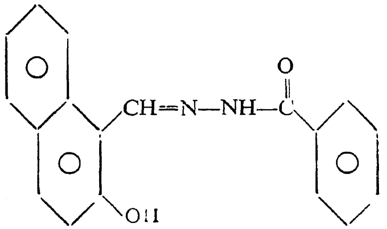
A few complexes of Ni (II) have been prepared by reacting nickel acetate with aroyl hydrazones in ethanol medium. The elemental analyses indicate 1 : 2 stoichiometry. The magnetic and electronic spectral data suggest an octahedral configuration for these complexes. The IR data show that the hydrazones coordinate through the oxygen of the carbonyl group and nitrogen of the azomethine group, suggesting that the hydrazones exist in the *keto form* in these complexes.

INTRODUCTION

In continuation of our previous work[1,2] on hydrazone complexes with transition metals, we now report the complexes of hydrazones with nickel. The hydrazones used are shown below.



I R = H
II R = CH_3
III R = Cl



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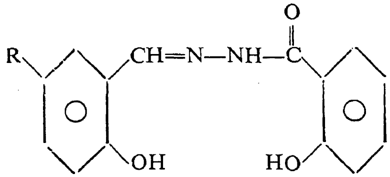
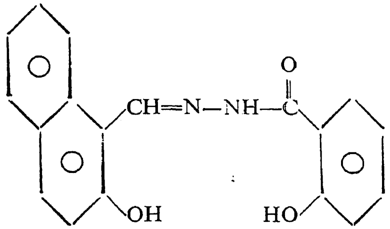


V R = H
VI R = CH_3
VII R = Cl



VIII

EXPERIMENTAL

Preparation of the Complexes

To an ethanolic solution containing 0·2 mol. of hydrazide and aldehyde, an aqueous solution of 0·1 mol. nickel acetate was added. The reaction mixture was heated for a while on water-bath and the complex precipitated was filtered and washed free from the ligand with ethanol and lastly with ether and dried in vacuum over P_2O_5. Nickel complexes were analysed for Ni, C, H and N, contents by standard methods.[3]

RESULTS AND DISCUSSION

The complexes are yellow in colour, insoluble in common organic solvents. However, they are soluble to a limited extent in DMF and DMSO. The results of the elemental analysis (Table I) show that these complexes have 1 : 2 stoichiometry.

Conductance

The molar conductance in DMF at concentration $\sim 10^{-3}$ M is too small to measure. Hence the complexes are non-electrolytes.

Magnetic Properties

The magnetic moments obtained at room temperature are in the range of 2·80 to 3·30 B.M. It has been well established that the Ni (II) complexes having octahedral configuration show magnetic moments in the range of 2·4 to 3·8 B.M.[4] These observations make us to conclude that Ni (II) has octahedral configuration in these complexes.

Electronic Spectra

It is known that the octahedral Ni (II) complexes exhibit three bands in the range 400–1200 nm attributable to the various *d–d* transitions[4]. In these complexes, we observe two broad bands in the region 675–860 nm and 960–1000 nm. The third band in these complexes is observed in the region 400–455 nm. Amongst these three bands, the band in the region 970–1000 nm is assigned to the $3A_{2g} \rightarrow 3T_{2g}$, transition. The band in the region 400–455 nm may be due to the $3A_{2g} \rightarrow 3T_{1g}$ (P). The middle band in the region 600–800 nm shows splitting due to spin orbit coupling and is assigned to the $3A_{2g} \rightarrow 3T_{1g}$ (F) transition. These observations suggest that Ni (II) in the complexes has an octahedral sp^3d^2 configuration. These observationss upport the conclusions adduced with the help of magnetic data.

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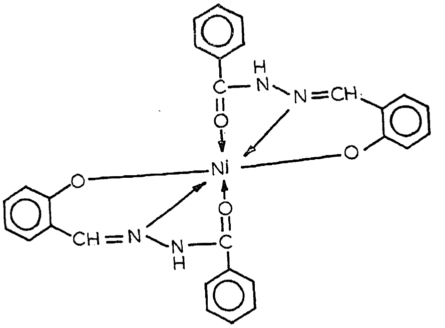
* For correspondence.

TABLE I

Elemental analysis, magnetic moment and electronic spectral data for Ni (II) complexes of hydrazones

Sl. No.	Ligand No.	Complex No.	Empirical formula	% Ni	% N	% C	% H	μ_{eff} (298° K), B.M.	Electronic spectra λ_{max} (nm)
1.	I	IX	$(C_{14}H_{11}N_2O_2)_2$ Ni	10·60 (10·93)	10·01 (10·43)	62·13 (62·61)	3·68 (4·09)	3·13	*430, 800, 1000
2.	II	X	$(C_{15}H_{13}N_2O_2)_2$ Ni	10·02 (10·40)	9·86 (9·92)	66·04 (67·08)	5·09 (4·60)	3·20	420, 800, 960
3.	III	XI	$(C_{14}H_{10}N_2O_2Cl)_2$ Ni	9·20 (9·69)	8·87 (9·25)	54·72 (55·47)	3·49 (3·30)	3·24	415, 675, 820, 970
4.	IV	XII	$(C_{18}H_{13}N_2O_2)_2$ Ni	8·69 (9·22)	9·02 (8·79)	67·98 (67·85)	4·55 (4·08)	3·15	*455, 700, 800, 1000
5.	V	XIII	$(C_{14}H_{11}N_2O_3)_2$ Ni	9·93 (10·32)	9·86 (9·85)	60·63 (59·08)	3·70 (3·87)	3·13	400, 700, 825, 970
6.	VI	XIV	$(C_{15}H_{13}N_2O_3)_2$ Ni	9·17 (9·84)	9·42 (9·39)	61·83 (60·33)	4·77 (4·36)	3·14	410, 700, 815, 970
7.	VII	XV	$(C_{14}H_{10}N_2O_3Cl)_2$ Ni	8·56 (9·21)	9·02 (8·78)	53·91 (52·69)	3·86 (3·14)	2·80	410, 695, 860, 970
8.	VIII	XVI	$(C_{18}H_{13}N_2O_3)_2$ Ni	8·54 (8·78)	8·84 (8·37)	64·73 (64·60)	3·60 (3·88)	3·30	445, 700, 970

Values in parentheses are the calculated values. * Electronic spectra in Nujol.



Infrared Spectra

The hydrazones show medium intensity band around 3200 cm^{-1} due to μ (NH)^5 vibration and is retained in the complexes. The broad weak band with fine structure due to intramolecular hydrogen bonded OH^6 in the region 2700–2600 cm^{-1} is not visualized in the complexes. The high intensity band in the region 1670-1650 cm^{-1} representing the C = O vibration^7 and that in the region 1625–1610 cm^{-1} due to C = N stretch^8 show considerable shift towards lower frequency in the complexes and appear in the regions 1640–1618 cm^{-1} and 1618–1602 cm^{-1} respectively. This may be interpreted as due to the coordination of C = O and C = N groups through oxygen and nitrogen atoms. The phenolic C—O around 1280 cm^{-1} appears in the region 1340-1300 cm^{-1}. This leads to the conclusion that the *o*-hydroxy group of the aldehyde residue has taken part in the complex formation.^{8}

These observations clearly show that in these complexes the ligands exist in the *keto form* and both the types of ligands show monovalent tridentate behaviour.

The analytical data show 1 : 2 stoichiometry. Magnetic and optical spectral results indicate agreement with the octahedral sp^{3}d^2 configuration of Ni (II) in the complexes. The infrared results indicate the monovalent tridentate behaviour of the ligands in these complexes.

All these observations make us to propose the following structure in which Ni (II) is hexacoordinated.

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1. Biradar, N. S., Mahale, V. B. and Havinale, B. R., *Curr. Sci.*, 1976, **45**, 6.
2. — and Havinale, B. R., *Inorg. Chim. Acta*, 1976, **17**, 157.
3. Vogel, A. I., *A Text Book of Quantitative Inorganic Analysis*, Longmans Green, London, 1969.
4. Cotton, F. A. and Wilkinson, G., *Advanced Inorganic Chemistry*, Wiley Eastern, New Delhi, 1972.
5. Rao, C. N. R., *Chemical Applications of Infrared Spectroscopy*, Academic Press, New York, London, 1963.
6. Freedman, H. H., *J. Am. Chem. Soc.*, 1961, **83**, 2900.
7. Martin, A. E., *Nature*, 1950, **166**, 474.
8. Biradar, N. S. and Kulkarni, V. H., *J. Inorg. Nucl. Chem.*, 1971, **33**, 2451.

SYMPOSIUM ON FERROUS AND FERRO-ALLOY MINERALS

The Department of Geology, Bangalore University, and the Post-graduate Centre, for Technical Studies, Nandihalli, Karnatak University, Dharwar, organised a Symposium on the problems of geology, beneficiation, exploration and metallurgy of ferrous and ferro-alloy minerals. The organisations like N.M.D.C., G.S.I., SMIORE, N.M.L. and UGC were the co-sponsorers of the symposium. The symposium was held in two sessions from 25th to 26th May 1976 at Bangalore and from 28th to 29th May 1976 at Sandur. More than 150 scientists from different organisations participated in the symposium.

On 25th May 1976, two technical sessions were held under the Presidentship of Prof. C. S. Pichamuthu and Prof. M. N. Viswanathaiah respectively. During these two sessions 11 papers were presented, dealing with the origin and enrichment of ferrous and ferro-alloy minerals (R. N. Mishra ; S. Acharya), their importance as marker horizons to decipher the stratigraphy of the region (C. Naganna and B. S. Shivakumar), and their occurrence in early precambrians and its significance (C. Naganna and B. Mahabaleswar).

On 26th, three technical sessions were held under the Chairmanship of Prof. M. S. Sadashivaiah, Dr. A. K. Chaterjee and Prof. O. P. Varma. The discussions focussed the attention on the following issues :

Mineragraphic studies on titanium and vanadium minerals and their paragenesis (C. Naganna *et al.*, N. Chayapathi, S. K. L. Rao), the structure and localisation of the mineral deposits (R. L. N. Dixit), Tungsten metallogenesis and its relation to the ferrous and ferro-alloy minerals (O. P. Varma and V. K. Singh), Crystallochemical and geochemical problems (K. T. Damodaran).

On 28th, two technical sessions were held at Nandihalli. Dr. M. V. N. Murthy and Sri Mahanti of Hutti Gold Mines chaired the sessions. The deliberations focussed the attention on the following points : 1. Better methods of exploration and exploitation (A. K. Chaterjee ; R. L. N. Dixit ; N. Acharya ; T. K. Bhattacharya) ; Resource estimation (T. Thimmaiah and R. H. Sawkar ; G. D. Kalra) and prospecting strategy (R. Srinivasan and B. L. Srinivas), 2. Different methods of beneficiation for low grade ferrous and ferro-alloy minerals (P. D. Prasada Rao *et al.*, P. K. Sinha *et al.*, K. T. Damodaran *et al.* and K. A. Natarajan), 3. Use of hydrocyclones as classification and concentration devices in both ferrous and ferro-alloy mineral industry (T. C. Rao).

On 29th May 1976, three technical sessions were held. Papers concerned with metallurgy of ferrous and ferro-alloy minerals were presented under the Chairmanship of Sri. S. Y. Ghorpade, G. V. Subramanyam and Sri. Pitre respectively. The attention was focussed on the following issues of vital importance : 1. Suitability of raw material for metallurgical industries (R. S. Rohidekar), 2. Design aspects of pellitisation system (D. N. Dey and A. K. Juhari), 3. Smelting of high carbon and low carbon ferromanganese in submerged arc electric furnace and arc furnaces (James D'Souza), 4. Effects of raw materials on the consistency of sinter strength and analysis (A. N. Mitra and T. Mukherjee), 5. Production of high purity magnetic concentrate by froth flotation using ionic collector oelic acid (M. G. Hebsur and N. Venkataraman), 6. Secondary refining processes and producing quality steels (N. D. Sreenivasa and K. P. Abraham). An exhibition of maps and minerals were arranged both at Bangalore and Nandihalli.

The first session of the symposium at Bangalore was inaugurated by Sri S. M. Krishna, Hon'ble Minister, for Industries and Parliamentary affairs and the second session of the symposium was inaugurated at Nandihalli, by Sri. M. Y. Ghorpade, Hon'ble Minister for Finance.

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LETTERS TO THE EDITOR

THERMODYNAMIC PROPERTIES OF SOME CHROMYL HALIDES

ONCE the spectroscopic measured frequencies and the moments of inertia of a polyatomic molecule are known it is possible to obtain the thermodynamical properties of the molecule. These properties have great practical importance since it is difficult and, sometimes, even impossible to measure these quantities experimentally. In certain cases the calculated values are more accurate than the experimental values. The reliability of the values of the force fields of vibrating molecules may be tested using the thermodynamical properties of the molecule.

TABLE I

Thermodynamic properties for CrO_2Cl_2 and CrO_2F_2

T (° K)	C_p° *	S° *	$\frac{H^\circ - E^\circ *}{T}$	$\frac{F^\circ - E^\circ *}{T}$
CrO_2Cl_2				
500	22·9680	91·2118	17·3919	73·8199
600	23·6893	95·4675	18·3854	77·0822
700	24·1806	99·1584	19·1803	79·9781
800	24·5262	102·4112	19·8281	82·5830
900	24·7765	105·3151	20·3648	84·9504
1000	24·9630	107·9357	20·8157	87·1200
1100	25·1050	110·3220	21·1995	89·1224
1200	25·2154	112·5113	21·5297	90·9815
1300	25·3029	114·5331	21·8167	92·7164
1400	25·3732	116·4109	22·0683	94·3426
1500	25·4306	118·1635	22·2906	95·8729
CrO_2F_2				
300	18·5420	75·2460	13·1083	62·1378
400	20·6394	80·8871	14·7459	66·1412
500	22·0444	85·6541	16·0746	69·5795
600	22·9875	89·7622	17·1532	72·6090
700	23·6351	93·3573	18·0357	75·3216
800	24·0925	96·5449	18·7658	77·7791
900	24·4248	99·4028	19·3771	80·0257
1000	24·6726	101·9396	19·8948	82·0948
1100	24·8615	104·3504	20·3381	84·0123
1200	25·0036	106·5202	20·7214	85·7988
1300	25·1251	108·5267	21·0559	87·4709
1400	25·2189	110·3923	21·3500	89·0423
1500	25·2954	112·1349	21·6106	90·5243

* Cal deg^{-1} $mole^{-1}$.

From the vibrational assignments[1-3] of the chromyl halides CrO_2X_2 (X = Cl or F) under consideration, it is found that they belong to the C_{2v} point group with symmetry number as one. The thermodynamic quantities, *viz.*, heat content, heat capacity, entropy and free energy, are calculated on the basis of the ground electronic state with rigid rotator harmonic oscillator approximation[5] and negligibly poor nuclear spin energy contribution. The results for the ideal gas state at atmospheric pressure are presented in Table I. From the structural parameters[2,4] of the molecules, the principal moments of inertia are calculated and presented in Table II.

TABLE II

Principal moments of inertia

Molecule	I_{xx}**	I_{yy}**	I_{zz}**
CrO_2Cl_2	330·83	158·67	271·84
CrO_2F_2	173·96	149·57	123·74

**a.m.u.Å².

The authors wish to thank Dr. K. N. Kuchela and Prof. M. A. Venkatachar, for the encouragement given to this work.

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---

1. Hobbs, N. E., *J. Chem. Phy.*, 1958, **28**, 1220.
2. Stammereich, K. *et al.*, *Spectrochim. Acta*, 1959, **15**, 438.
3. Miller, G. *et al.*. *Ibid.*, 1959, **15**, 709.
4. Kerroknox, *Acta Cryst.*, 1960, **13**, 507.
5. Herzberg, G., *Molecular Spectra and Molecular Structure*—II.

THIORIDAZINE HYDROCHLORIDE AS A NEW REAGENT FOR THE SPECTROPHOTOMETRIC DETERMINATION OF OSMIUM (VIII)

Introduction

DURING the spectrophotometric study of noble metals using thioridazine hydrochloride (TH)[1-3], it was found that TH formed blue-coloured oxidation product with osmium (VIII) in hydrochloric acid medium. This reaction is now utilised for the spectrophotometric determination of osmium (VIII).

Experimental

Stock solution of osmium (VIII) was prepared from osmium tetroxide in 0·2 M sodium hydroxide and standardised as described by Ayres and Wells[4]. Working solutions were prepared as needed by the dilution of the stock solution. A 0·2% of aqueous solution of TH was used. All other solutions were prepared from reagent grade chemicals. Beckman Model DB spectrophotometer with matched 1 cm silica cells was used for absorbance measurements.

Procedure.—An aliquot of the sample solution containing 2·5–72 ppm of osmium was transferred to a 25 ml volumetric flask. 5 ml of 5 M hydrochloric acid and 5 ml of 0·2% TH solution were added and diluted to the mark with doubly distilled water. The solution was thoroughly mixed and the absorbance was measured after 10 minutes at 640 nm against a reagent blank prepared in the same way. The osmium concentration of the sample was calculated by reference to a previously prepared calibration curve.

Results and Discussion

Osmium (VIII) oxidises TH to a blue-coloured product which is thought to be a free radical[3]. The sensitivity and the stability of the blue-coloured product depend on the nature and strength of acid medium. The sensitivity of the reaction in four acid media (1·0 M) is in the order, HCl > H_2SO_4 > H_3PO_4 > HAC. The stability of the blue colour in 1·0 M HCl, H_2SO_4, H_3PO_4 and HAC is 25, 10, 5 and 4 minutes respectively. Hence hydrochloric acid medium is preferred. The maximum colour intensity is observed in 0·2–1·5 M HCl. Therefore 1·0 M hydrochloric acid is used in these studies.

The electronic spectrum of the blue-coloured species shows the maximum absorption around 640 nm with a shoulder at 490 nm. The reagent does not absorb at these wavelengths. A thirty-fold molar excess of TH over osmium is required for complete colour development. The constant absorbance values are obtained 5 minutes after adding the reagent to the osmium solution. The order of addition of reagents has no effect on the absorbance values which are insensitive to temperature in the range 5–40° C.

Beer's law is valid within the concentration range of 0·1 to 2·8 ppm of osmium and the optimum range is 1·0 to 2·4 ppm of osmium. At 640 nm, the molar absorptivity is $1{\cdot}98 \times 10^4$ litre mole^{-1} cm^{-1}. For log $I_0/I = 0{\cdot}001$, the sensitivity of the reaction is 0·0096 μg Os cm^{-2}.

In order to assess the possible analytical applications of the reaction, the effect of diverse ions was studied. The following amounts (μg/ml) of foreign ions were found to give less than 2% error in the determination of 2 ppm of osmium; Pt (IV) 15; Rh (III) 12; Ru (III) 1; Ir (III) 15; Co (II) 100; Ni (II) 150; UO_2 (II) 100; fluoride 1100; chloride 1800; bromide 800; nitrate 900; sulphate 1000; phosphate 300; oxalate 850; EDTA 10. Iodide, thiosulphate, Pd (II), Au (III), Fe (III), Ag (I), Ce (IV) and V (V) interfere seriously. Attempts to increase the tolerance limits of some cations were unsuccessful.

Determination of osmium in ores.—Synthetic mixtures containing platinum metals corresponding to osmiridium or syserkite were prepared and the osmium content was determined. The results are given in Table I.

TABLE I

Determination of osmium in synthetic mixtures corresponding to osmiridium or syserkite

Osmium present ppm	Rh, ppm	Ir, ppm	Ru, ppm	Pd ppm	Pt, ppm	Osmium found, ppm
1·00	..	0·250	0·150	..	0·050	1·02
1·50	..	0·425	0·225	..	0·075	1·56
2·00	..	0·500	0·300	..	0·100	2·05

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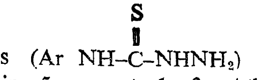
1. Sanke Gowda, H. and Ramappa, P. G., *Curr. Sci.*, 1976, 45, 218.
2. —. — and Narayana Achar, B., *Z. anal. Chem.*, 1975, 275, 127.
3. — and —, *Ibid.*, 1976, 280, 221.
4. Ayres, G. H. and Wells, W. N., *Anal. Chem.*, 1950, 22, 317.

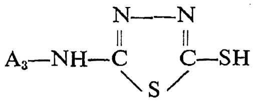
STUDIES IN THIADIAZOLES: SYNTHESIS OF SOME 2-SUBSTITUTED-5-MERCAPTO-1, 3, 4-THIADIAZOLES



4-ARYLTHIOSEMICARBAZIDES ($\mathrm{Ar\ NH{-}\overset{\displaystyle S}{\overset{\|}{C}}{-}NHNH_2}$) on cyclisation yield 2-arylamino-5-mercapto-1, 3, 4-thiadiazoles which have been found to be biologically active[1]. The SH group is an important toxophore as —OH especially to fungi[2] and when a heterocyclic compound contains an —SH group adjacent to a nitrogen atom, useful fungitoxic compounds are often obtained because of the possibility of chelation[2,3]. The 2-arylamino-5-mercapto-1, 3, 4-thiadiazoles fulfil these requirements and therefore a study of these compounds was undertaken with a view to studying their fungicidal activity.

In the present communication six thiadiazoles have been prepared by the reaction of 4-arylthiosemicarbazides with carbon disulphide and diemethylformamide[4]. (i) 4-*Arylthiosemicarbazides* were prepared

TABLE I



Sl. No.	Ar.	Yield	M.P. °C	Mol. formula	% Sulphur Found	% Sulphur Reqd.	Dia. of zone of inh. in mm
1.	4-Iodo-2-methylphenyl	60	184	$C_9H_8N_3S_2I_2$	18·46	18·34	..
2.	2, 5-Dimethoxyphenyl	70	204	$C_{10}H_{11}O_2N_3S_2$	23·66	23·79	10
3.	2, 5-Dichlorophenyl	65	130	$C_8H_5N_3S_2Cl_2$	23·16	23·02	12
4.	2-Bromo-4-methylphenyl	68	186	$C_9H_8N_3S_2Br$	21·32	21·19	..
5.	4-Chloro-2-methoxyphenyl	62	172	$C_9H_8N_3OS_2Cl$	23·58	23·40	11
6.	4-Chloro-2-methylphenyl	64	182	$C_9H_8N_3S_2Cl$	24·72	24·85	10
	Benzoic acid						13

by the method of Kazokov[5]. (ii) 2-*Arylamino*-5-*mercapto*-1, 3, 4-*thiadiazoles*: A mixture of appropriate 4-arylthiosemicarbazide (0·01 M), carbon disulphied (0·01 M) and dimethylformamide (20 ml) was refluxed on the water bath for about 1–1·5 hours at 65–70° C. The excess of the solvent was distilled off under reduced pressure. On cooling, the product separated out and was purified.

The details of the compounds thus prepared are listed in Table I.

Pharmacology: The prepared thiadiazoles were tested against five strains of fungi, *viz*., *C. albicans*, *C. utilis*, *A. tenuis*, *A. niger* and *A. flavus* for fungicidal actvity at a concentration of 150 ± 10 μg/ml in 50% ethanol. The strength is reported by measuring the diameter of zone of inhibition of a particular microorganism (Table I) and the results were compared against beznoic acid.

The authors wish to record their sincere thanks to Dr. R. S. Kapil, Central Drug Research Institute, Lucknow and Dr. S. K. Singhal for the assistance during the course of this work.

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1. *U.S. Patent*, 1950, **825**, 2497; *C. Abstr.*, 1950, **44**, 5919.
2. Horsfall and Rich, *Contribs Thompson Inst.*, 1951, **16**, 313.
3. Albert *et al.*, *Brit. Jour. Expt. Path.*, 1947, **28**, 69.
4. Giri, S. and Singh, H., *J. Indian Chem. Soc.*, 1967, **44**, 145.
5. Kazokov, V. Ya. *et al.*, *C. Abstr.*, 1961, **55**, 23415.

GENETICS AND BIOCHEMISTRY OF PYRIDOXINE REQUIRING MUTANTS OF *ASPERGILLUS NIDULANS*

IN our attempts to isolate a number of nutritionally deficient mutant strains of *Aspergillus nidulans* by ultraviolet irradiation of spores from a parent strain *y* ; $ribo_1$ (spore colour yellow and requiring riboflavin for growth), eight pyridoxine requiring mutants were obtained. Seven of them were found to be non-allelic by complementation tests[1]. The mutants were outcrossed with a strain *bi* ; w_3, ade_3 (white spore colour requiring both biotin and adenine for growth). Strains requiring only pyridoxine for growth were isolated from the perithecia and designated as $pyro_5$, $pyro_6$, $pyro_7$, $pyro_8$, $pyro_9$, $pyro_{10}$ and $pyro_{12}$, with the spore colours white, yellow which are recessive and the dominant green. Since all the mutant strains were available with white spore colours, excepting $pyro_8$, strains with white conidial colours were taken up for the present study, along with $pyro_8$ with yellow conidial colour.

The pyridoxineless mutants were allowed to grow at 23° C, 30° C, 37° C and 42° C on pyridoxine supplemented Minimal Medium[1] and were found to be temperature independent. The growth promoting effect of sugar phosphates, three carbon compounds of the glycolytic pathway, purines, pyrimidines, nicotinic acid and some other alleged precursors of pyridoxine are presented in Table I. From the results presented, it can be seen that β-glycerophosphate, dihydroxy acetone phosphate, 2- and 3-phosphoglycerates could promote the growth of mutant strain w_3 ; $pyro_9$. The mutant

TABLE I

Effects of compounds related to pyridoxine or conceivably involved in its biosynthesis added in the absence of pyrioxine. Substances added at a level of 800 *µg*/10 *ml of medium in* 50 *ml Erlenmeyer's flasks. Mycelial weights were obtained from* 10 *ml cultures after growth for* 7 *days. Results expressed in mg* ± *S.D.*

Compounds tested	Strains tested						
	$w_3; pyro_5$	$w_3; pyro_6$	$w_3; pyro_7$	$y_1; pyro_8$	$w_3; pyro_9$	$w_3; pyro_{10}$	$w_3; pyro_{12}$
1. Minimal Medium alone	—	—	—	—	—	—	—
2. MM + all amino acids individually	—	—	—	—	—	—	—
3. MM + Purines	—	—	—	—	—	—	—
4. MM + Pyrimidines	—	—	—	—	—	—	—
5. MM + Glucose-6-phosphate	—	—	—	—	—	—	—
6. MM + Fructose-6-phosphate	—	—	—	—	—	—	—
7. MM + Fructose-1, 6 diphosphate	—	—	—	—	—	—	—
8. MM + β-glycerophosphate	—	—	—	—	35·0±1·5	—	—
9. MM + Dihydroxy acetone phosphate	—	—	—	—	32·0±1·5	—	—
10. MM + 3-Phosphoglyceric acid	—	—	25·0±2·2	—	25·5±1·7	—	—
11. MM + 2-Phosphoglyceric acid	—	—	29·5±3·0	—	33·0±1·4	—	—
12. MM + 3-Phosphoserine	—	—	33·5±3·9	—	—	37·5±2·7	—
13. MM + Phosphoenolpyruvate	—	—	35·0±2·5	—	—	—	—
14. MM + Pyruvate	—	—	34·0±1·5	—	—	—	—
15. MM + Kynurenine	—	—	—	33·5±1·7	—	—	—
16. MM + Formyl kynurenine	—	—	—	34·0±1·8	—	—	—
17. MM + 3-hydroxy anthranilic acid	—	—	—	38·0±1·6	—	—	—
18. MM + Nicotinic acid	—	—	—	42·5±4·1	—	—	—
19. MM + Pyridoxine	36·0±1·5	44·0±2·3	45·0±2·5	42·5±1·5	38·1±1·5	38·5±1·5	30·0±1·

— Indicates inoculum weight too small to measure.

strain w_3; $pyro_7$ could grow in the presence of 2- and 3-phosphoglycerates, pyruvate and 3-phosphoserine. Only 3-phosphoserine could promote the growth of w_3; $pyro_{10}$. These results indicate that β-glycerophosphate, dihydroxy acetone phosphate, 2- and 3-phosphoglycerates, pyruvate and 3-phosphoserine are growth promoting for some of the pyridoxine requiring mutants of *Aspergillus nidulans* and thus they may be involved in the biosynthesis of pyridoxine. It is significant to note that only the phosphoesters showed growth promoting activity, while glucose, fructose, glyceric acid, glycerol and serine did not promote growth for the respective mutant strains. This proves that the phosphoesters are active directly and not after hydrolysis (as could happen in the presence of phosphatase in culture filtrate).

Earlier observations made on a pyridoxine requiring mutant of *E. coli* by radioactive incorporation studies using isotopes of dihydroxy acetone phosphate and glyceraldehyde-3-phosphate[2,3,4,5] indicated that C-5, C-5′ and C-6 of pyridoxine could be derived from glyceraldehyde-3-phosphate and C-3, C-4 and C-4′ of pyridoxine from dihydroxy acetone phosphate, thus showing that the three carbon metabolic units in the glycolytic intermediates can serve as the source of cyclic precursors for the pyridoxine molecule. A series of studies made on a pyridoxine requiring mutant of *E. coli*[6,7,8] indicated that 3-phosphoserine could be a possible precursor of pyridoxine. These observations agree well with our observations.

One of the interesting observations made by us is that kynurenine, formyl kynurenine, 3-hydroxy anthranilic acid and nicotinic acid (which is a pyridine derivative like pyridoxine) are able to promote the growth of the mutant y_1; $pyro_8$. Neither amino acids, pyrimidines, purines nor sugar phosphates show any growth promoting activity in the place of pyridoxine.

The role of three carbon compounds and nicotinic acid in the biosynthesis of pyridoxine are being investigated in detail.

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---

1. Pontecorvo, G., Roper, J. A., Hemmons, L. M., MacDonald, K. D., and Bufton, A. W. J., *Adv. in Genetics*, 1953, 5, 141.
2. Hill, R. E. and Spenser, I. D., *Science*, 1970, 169, 773.
3. — and—, *J. Amer. Chem. Soc.*, 1971, 93, 518.
4. —, Rowell, F. J., Gupta, R. N. and Spenser, I. D., *J. Biol. Chem.*, 1972, 247, 1869.
5. — and Spenser, I. D., *Can. J. Biochem.*, 1973, 51, 1412.
6. Dempsey, W. B., *J. Bacterial.*, 1969, 97, 1403.
7. —, *Biochem. Biophys. Res. Commun.*, 1969, 37, 89.
8. — and Itoh, H., *J. Bacteriol*,. 1970, 104, 658.

IN VITRO TOXICITY OF CONSTITUENTS OF *RUMEX MARITIMUS* LINN. TO RINGWORM FUNGI

SEVERAL species of the genus *Rumex* Linn. (N.O. Polygonaceae) are used medicinally in Europe, South Africa and Madagascar, for various ailments including as an antidote to poisonous insect bites and cutaneous disorders[1]. In the screening of Indian plants for biological activity, alcoholic extract of *R. maritimus* Linn. was shown to exhibit activity *in vitro* against the two ringworm fungi, *Trichophyton mentagrophytes* and *Microsporum canis* for the first time[2] which prompted the present investigations.

Isolation and identification of constituents.—The alcoholic extract of the whole plant was fractionated into hexane and butanol-soluble fractions. The hexane-soluble residue was chromatographed over silica gel and three crystalline substances (A, B and C) were isolated. The butanol-soluble fraction on chromatography over silica gel yielded a crystalline substance D. *Substance A*, pale yellow flakes from chloroform-alcohol, m.p. 204°, $C_{16}H_{12}O_5$. A magenta colour with sulphuric acid and red-brown colour with ferric chloride indicated it to be an anthraquinone. $\nu_{max}^{(KBr)}$: 3350, 1040 (OH), 1680 (free carbonyl), 1620 (chelated carbonyl) and 1560, 755 cm^{-1} (aromatic). λ_{max} : 226, 255, 268, 289 and 440 nm (log ϵ 4·43, 4·21, 4·24, 4·29 and 4·05). MS : m/e 284 (M^+).

It formed a diacetate, m.p. 182–4°. $C_{20}H_{16}O_7$ (M^+, 368), monomethylether, mp. 187–9°, $C_{17}H_{14}O_5$ (M^+, 298), dimethylether, mp. 221–4°. $C_{18}H_{16}O_5$ (M^+, 312) and was identified as physcion. *Substance B.* m.p. 137°, $(\alpha)_D$ — 35°. Monoacetate, mp. 126–7°. It was confirmed as β-sitosterol.

Substance C, yellow needles from chloroform-alcohol, m.p. 252–3°, $C_{15}H_{10}O_5$. It also gave a magenta colour with sulphuric acid and red-brown colour with ferric chloride. $\nu_{max}^{(KBr)}$: 3500, 1040, 1670, 1630, 1600, 765 and 730 cm^{-1}. λ_{max} : 223, 253, 266, 289 and 439 nm (log ϵ 4·55, 4·26, 4·23, 4·29 and 4·10). MS : m/e 270 (M^+).

It yielded a triacetate, m.p. 194–7°, $C_{21}H_{16}O_8$ (M^+, 396). With diazomethane, it formed a monomethylether, mp. 203–4°, $C_{16}H_{12}O_5$ (M^+, 284) which was identical with physcion. This established substance C as emodin.

Substance D, needles from methanol, mp. 280–4°. $(\alpha)_D$ — 44°, $C_{35}H_{60}O_6$. It gave positive Fiegel's test. Tetraacetate, m.p. 170°. On hydrolysis with 6.N HCl, it yielded β-sitosterol, and glucose which confirmed it as β-sitosterol-β-D-glucoside.

Toxicity to ringworm fungi.—Fragments of hyphae of the ringworm fungi, grown on Sabouraud's dextrose agar slants[3] for fifteen days, were introduced into Erlenmeyer flasks containing 50 ml Sabouraud's dextrose broth to which 0·004% agar was added. The flasks were incubated at 28 ± 1° on shaker for 48 hr., the clear broth was dispensed in assay tubes and two fold serial dilution method was used for testing the activity. The test materials were dissolved in alcohol to obtain a 10 mg/ml solution. Suitable solvent and test fungi controls were maintained. Observations were made after 96 hr. incubation at 28 ± 1°. The minimum inhibitory concentration (MIC) was calculated from the tube showing no visible growth. The effect of solvent in inhibiting the growth of test fungi was observed only in first two tubes and hence the plant material inhibiting the growth at a concentration of 250 μg/ml (third tube) was considered active. The maximum concentration of the plant materials tested was 1000 μg/ml and none of them altered the pH of the test broth (pH 5·8–6·0). The results are presented in Table I.

TABLE I

Antifungal activity of plant materials

Plant materials	MIC (μg/ml) against:	
	T. mentagrophytes	*M. canis*
Crude alcoholic extract	250·00	125·00
Hexane-soluble fraction	62·50	31·25
Butanol-soluble fraction	125·00	62·50
Aqueous fraction	Inactive	Inactive
Physcion	,,	,,
β-sitosterol and its glucoside	,,	,,
Emodin	250·00	125·00

R. maritimus Linn. has been reported to contain rutin, hyperin, rumarin, chrysophanol, physcion and emodin[4]. Emodic acid, aloe-emodin, rhein and rhein-like substances have been found only in fruits[5]. We have found only physcion, β-sitosterol, its glucoside and emodin in the Indian species investigated by us.

The crude alcoholic extract, hexane and butanol fractions and emodin from *R. maritimus* Linn. have been shown in this study to be toxic to ringworm fungi.

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1. Kirtikar, K. R. and Basu, B. D., *Indian Medicinal Plants*, Lalitmohan Basu, Allahabad, 1933, 3.
2. Dhar, M. L., Dhar, M. M., Dhawan, B. N., Mehrotra, B. N., Srimal, R. C. and Tandon, J. S., *Indian J. exp. Biol.*, 1973, 11, 43.
3. Srivastava, O. P. and Gupta, R. N., *J. Sci. Industr. Res.*, 1958, 17 C, 87.
4. Bagrii, A. K., Kurmaz, B. V. and Litvinenko, V. I., *Khim. Prirodn. Soedin.*, 1966, 2, 85.
5. Fairbairn. J. W. and El-Muhtadi, F. J., *Phytochemistry*, 1972, 11, 263.
6. Walker, J. C., Morrel, S. and Foster, H. H., *Amer. J. Bot.*, 1937, 24, 536.

MANGO PEEL WASTE AS A SOURCE OF PECTIN

MANGO peel which constitutes 20–25% of the total weight of the fruit was suggested as cattle feed[1]. However, no organised efforts have so far been made for its effective utilization. Pectin, a biological polymer containing partially methylated galacturonic acid is recognised as an indispensable ingredient in the food processing industry for the manufacture of jams, jellies and as a thickner in a variety of foods and in pharmaceutical preparations[2]. An attempt was therefore made to extract pectin from mango peel and ascertain its physico-chemical properties. The characteristics of Alphonso mango peel pectin are shown in Table I which indicate that this is comparable with good quality apple pectin suitable for commercial exploitation. The average yield of mango peel pectin on dry weight basis was found to be 13% as compared to apple pomace with 17% yield[2] and mandarin orange peel with 13·2% yield[3,4].

TABLE I

The physical and chemical characteristics of mango peel pectin (Mangifera indica, *var. Alphonso*)

Yield (d.w.b.) %	13·0
Moisture %	4·30
Ash %	0·55
Methoxyl content %	8·20
Anhydrouronic acid %	61·12
Degree of Esterification %	76·0
Molecular weight	105,000
Equivalent weight	964
Optical rotation $(\alpha)_D^{25}$	+182°
Setting time (mins)	10
Jelly grade	200

The pectin from mango peel rates well in comparison to mandarin orange peel (Coorg and Nagpur varieties) which has been shown to contain pectin of 150–200 grade[3,4]. However, orange cultivation is localized to small regions while mango is grown more extensively all over India. This therefore points to the possible utilization of wastes from mango processing industry and also as a substitute to imported pectin preparations.

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---

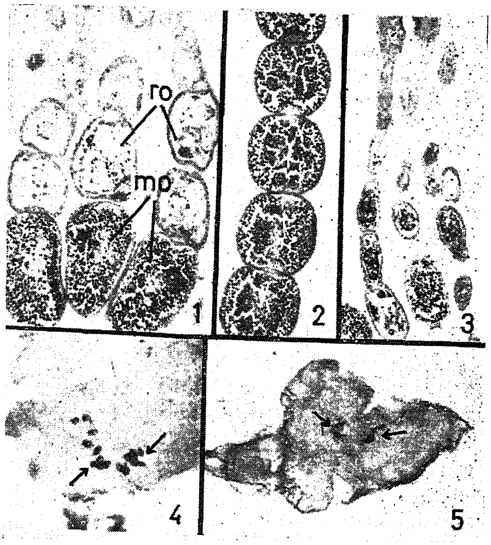
1. Pruthi, J. S., Krishnamurthy, G. V. and Lal, G., *Ind. Food Packer*, 1959, 13, 7.
2. Kertesz, Z. I., *The Pectic Substances*, Published by Interscience Publishers, Inc., New York and London, 1951, p. 628.
3. Pruthi, J. S., Parekh, C. M. and Lal, G., *Food Sci.*, 1961, 10, 372.
4. Agarwal, P. C. and Pruthi, J. S., *Ind. Food Packer*, 1972, 26 (2), 9.

ECDYSTERONE INDUCED OVARIAN INHIBITION IN THE COTTON BUG, *DYSDERCUS CINGULATUS*

ECDYSTERONE or some of its analogues inhibited vitellogenesis in the stable fly *Stomoxys calcitrans*[1,2], in the house fly *Musca domestica*[3,4], in the flour beetle *Tribolium confusum*[3], in the boll weevil *Anthonomus grandis*[5] and in the cockroach *Leucophaea maderae*[6]. However, these hormones stimulated vitellogenesis in the mosquito *Aedes aegypti* in larger doses, but smaller doses inhibited it[7]. So it is suggested that small doses of ecdysterone and analogues present in some plants perhaps serve to inhibit reproduction in plant feeding insects. Since very little evidence is available regarding the influence of ecdysone analogues on reproduction in the female plant bugs which could serve as ideal material for testing this hypothesis it is decided to study small doses of ecdysterone on reproduction in the female red cotton bug *Dysdercus cingulatus*.

Female *Dysdercus cingulatus*, a day after adult emergence were employed for the present study. They were injected ecdysterone (B-ecdysone) dissolved in distilled water on four consecutive days from day 1 onwards, either 2 μg or 4 μg doses after ether anethesia, through pleural region by means of a Hamilton microliter syringe. Animals receiving equal quantity of distilled water served as controls. The animals were kept along with some males in glass chimneys top covered with clothing. They were fed with cotton seeds soaked in water. After five days, the ovaries and brains of 10 females of each group were fixed in Bouin's fluid and were processed, sectioned and stained in iron alum haematoxylin eosin. Brains were stained as a whole using Aldehyde–Fuchsin or Victoria–Blue technique[8]. The remaining animals were kept for a couple of days more and were observed for mating and egg laying.

Mortality was high among animals treated with 4 μg dose. However, the surviving experimental animals were as healthy as controls and mated normally. Both the doses of ecdysterone inhibited normal development of the oocytes. In the injected animals only one to four oocytes in the basal region of each ovariole developed fully (Fig. 1) and such oocytes were comparable in all respects to the basal oocytes of the control animals. But in the controls 8–10 oocytes developed fully in each ovariole (Fig. 2). In the treated animals, oocytes anterior to the developing ones either resorbed or had no yolk granules, but were filled with



FIGS. 1–5. Fig. 1. Section of the ovary of animal 5 days after injection of ecdysterone showing the mature basal oocytes and the anterior resorbing oocytes with very few yolk granules. *mo*—mature oocytes; *ro*—resorbing oocytes, × 22·50. Fig. 2. Section of the ovariole of control of the same age as above showing mature oocytes, × 22·50. Fig. 3. Section of an ovary of an animal 5 days after injection of ecdysterone showing resorption of all oocytes. × 22·50. Figs. 4 and 5. Whole mounts of the brains of animals 5 days after injection of ecdysterone and its control respectively, showing increase in the material of the A-cells (arrows) in the injected animals, × 50.

vacuoles (Fig. 1). Their follicle cells were very small and degenerating or they invaded into the empty ovarian tubes. In some animals almost all oocytes resorbed (Fig. 3). In the germarium, disintegration of trophocytes was more intense in the treated animals. Insects which were kept for 6 days or more laid eggs though the number of eggs laid were only about 50% of those of the controls. Median neurosecretory cells of the brain in the treated insects (Fig. 4) showed pronounced accumulation of secretory material when compared to those of controls (Fig. 5) at the time of sacrifice.

The present studies on *Dysdercus cingulatus* showed that ecdysterone did not completely suppress vitellogenesis unlike in the stable fly[1] or in the weevil[5]. While the number of eggs in which vitellogenesis took place was drastically reduced to about 50% of those of the controls, vitellogenesis apparently proceeded normally in these eggs which were laid ultimately. However, the remaining eggs degenerated and resorbed. Hence it appeared that ecdysterone inhibited vitellogenesis in so far as the number of eggs undergoing vitellogenesis decreased whereas in the remaining eggs vitellogenesis was completely inhibited and they degenerated. It appears that small amounts of ecdysterone present in some plants might have an inhibiting influence on population density of the bugs as hypothesised by Spielman *et al.*[7], in the case of mosquitoes.

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---

1. Wright, J. E. and Kaplanis, J. E., *Ann. ent. Soc. Am.*, 1970, 63, 622.
2. —, Chamberlain, W. F. and Barrett, C. C., *Science*, 1971, 172, 1247.
3. Robbins, W. E., Kaplanis, J. N., Thompson, M. J., Shortino, T. J., Cohen, C. F. and Joyner, S. C., *Ibid.*, 1968, 161, 1158.
4. Rezabova, B., Hora, J., Landa, V., Cerny, V. and Sorm, F., *Steroids*. 1968, 11, 457.
5. Earle, N. W., Padovani, I., Thompson, M. J. and Robbins, W. E., *J. econ. Entomol.*, 1970, 63, 1064.
6. Engelmann, F., *Z. Vergl. Physiol.*, 1959, 41, 456.
7. Spielman, A., Gwadz, R. W. and Anderson, W. A., *J. Insect Physiol.*, 1971, 17, 1807.
8. Dogra, G. S. and Tandon, B. K., *Quart. J. micr. Sci.*, 1964, 105, 455.

INFLUENCE OF A JUVENILE HORMONE ANALOGUE* ON METAMORPHOSIS AND REPRODUCTION IN THE CRICKET, *PLEBEIOGRYLLUS GUTTIVENTRIS* WALKER

WIGGLESWORTH's classical work[1] on the role of corpora allata in growth, metamorphosis and reproduction of insects has been reinvestigated recently using a variety of compounds mimicking the juvenile hormones[2-8]. However, such an investigation has not been carried out on crickets. This paper deals with the effect of a juvenile hormone analogue (JHA) on metamorphosis and reproduction in the cricket, *Plebeiogryllus guttiventris.*

A total of 126 last instar nymphs from the stock culture[9] was selected for this experiment. One µl of JHA was topically applied with a micropipette to their last three abdominal tergites. This dosage was not only convenient for application but also showed considerable effectivity and least mortality. Time of JHA application varied from 0–168 h after moulting to last nymphal instar. Twenty nymphs were treated with 1 µl olive oil to serve as controls. Observations regarding moulting duration, metamorphic changes and reproductive abilities were recorded.

Based on the morphological features of nymphs and adults, four new forms (stages) were obtained for the JHA treatment (Fig. 1): I, supernumerary larva similar to 7th instar but with larger body size and curved wing pads; II, intermediate adult having more nymphal features but with incomplete and distorted adult wings and ovipositor;

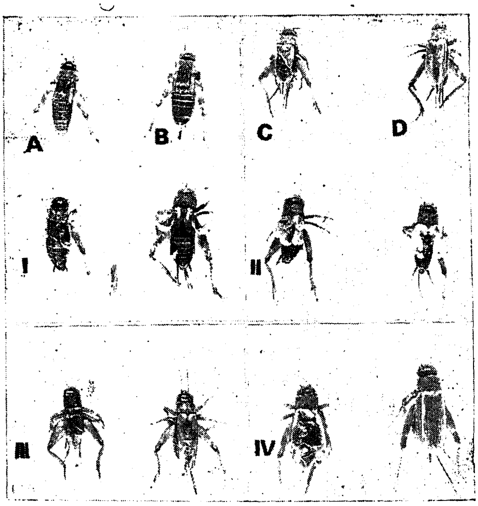

FIG. 1. (I), Supernumerary larvae; (II), intermediate adults; (III), adultoids and (IV) giant adults for the JHA application.

A and B: 7th instar control nymphs; C and D: Control adults.

III, adultoids with adult wings and ovipositor but with smaller body and pale colouration and IV, giant adults, moulted from the supernumerary larvae, with body size 1·6 times larger than control adults.

Time of JHA application influenced the degree of metamorphosis. Application, prior to 72 h after moulting, resulted in supernumerary larvae and thus completely inhibited metamorphosis. While percentages of surviving forms attaining stages I, II and III were 7, 40 and 53 for the nymphs treated between 72–96 h after the moult, they were 0, 13 and 87 for those treated between 96–120 h. JHA treatment after 120 h produced stage III only. All supernumerary larvae moulted to giant adults. Thus JHA action was critical and time dependent during the first one-third period of last nymphal instar. Later application led to decreasing juvenilisation with appearance of intermediate adults and adultoids. This observation conforms to the findings for *T. molitor*[4] and *R. prolixus*[6].

Further, JHA application influenced the duration of 7th nymphal instar. While these last instar nymphs took 8·16 ± 0·39 days to moult to 8th instar (supernumerary), control nymphs showed 9·15 ± 0·09 days to moult to normal adults. The difference was found to be statistically significant ($P < 0{\cdot}05$). Such accelerated moulting cycle because of JH action has also been recorded for *R. prolixus*[6] and *T. molitor*[10].

Intermediate adults failed to mate, nor did they deposit or produce any eggs. But adultoids mated normally and even produced and deposited eggs. Giant females, though mated with normal males, deposited only a few eggs. But they had retained many developed eggs in their body. Giant males, on the contrary, failed to mate as they could not produce any spermatophores. Their accessory glands were found to lack well developed tubules which might be the cause for their inability to produce spermatophores. Malformation of male genitalia and thereby failure to mate as a result of JHA treatment has been recorded for *D. fasciatus*[3].

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---

* RO-20-3600, Hoffmann-La Roche.

1. Wigglesworth, V. B., *Quart. J. Micr. Sci.*, 1937, 79, 91.
2. Mehrotra, K. N., Rao, P. J. and Phokela, A., *Indian J. Ent.*, 1969, 31, 183.
3. Critchley, B. R. and Campion, D. G., *Bull. Ent. Res.*, 1971 *a*, 61, 49.
4. — and —, *Ibid.*, 1971 *b*, 61, 293.
5. Brookes, V. J., *Israel J. Ent.*, 1972, 7, 85.
6. Barrett, M. F., *J. Insect Physiol.*, 1974, 20, 1507.
7. Silhacek, D. L. and Oberlander, H., *Ibid.*, 1975, 21, 153.
8. Das, Y. T. and Gupta, A. P., *Experientia*, 1974, 30, 1093.
9. Mathad, S. B. and Dakshayani, K., *Ann. Entomol. Soc. Amer.*, 1972, 65, 282.
10. Socha, R. and Sehnal, F., *J. Insect Physiol.*, 1973, 19, 1449.

ON THE SYNONYMY OF *GANEO KORKEI* BHALERAO, 1936 AND *GANEO PUNJABENSIS* GUPTA, 1954 WITH *GANEO TIGRINUM* MEHRA AND NEGI, 1928

THE genus *Ganeo* was erected by Klein, 1905, for the worms he collected from the intestine of *Rana hexadactyla* and designated as *Ganeo glottoides*. Subsequently a number of species were added to this genus, but in the present communication the authors are concerned with three species, *viz.*, *G. tigrinum* Mehra and Negi, 1928; *G. korkei* Bhalerao, 1936, parasitizing the intestine of *Rana tigrina* and *G. punjabensis* parasitizing the intestine of *Rana cyanophlyctis*.

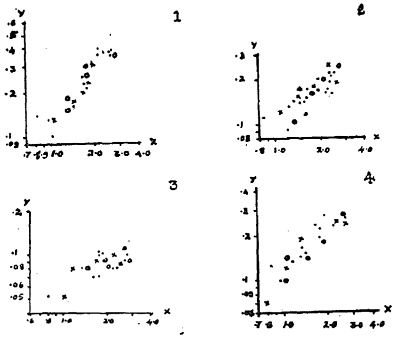
G. korkei and *G. punjabensis* differ from *G. glottoides* in the absence of pseudocirrus sac and in possessing a 'U'-shaped excretory bladder with a median stem, but in these respects they closely resemble *G. tigrinum*: however they differ from *G. tigrinum* in having the grouping of vitellaria. Further they are distinguishable from each other on the size of the gonads. In *G. korkei* ovary is larger than testes and in *G. punjabensis* ovary is smaller than testes.

The distinction between these two species does not hold good for tht fact that the size of the gonads is variable in *G. tigrinum*. An examination of a number of ten species of *G. tigrinum* showed that in various specimens of the same species ovary may be larger, equal or smaller than the testes.

When these variations between gonad sizes occur within one species they become intraspecific variations and all the species erected on this basis lose their identity. Hence, *G. punjabensis* becomes synonym of *G. korkei*. But the distinction of *G. punjabensis* from *G. tigrinum* also does not exist for the fact that in the *genus ganeo* the position and arrangement of vitellaria presents many variations, specially in those in which the pseudocirrus sac is absent. Hence this particular parameter is not of much taxonomic importance as discussed elsewhere (Rao, 1974) and as such *G. korkei* and *G. punjabensis* become synonym to *G. tigrinum.*

The synonymity of these two species with *G. tigrinum* is further supported by a study of allometric growth developed by Rohde (1966). According to him growth rate of an organ is much slower compared to the whole body and is expressed by the formula $\log y = \log b + a . \log x$, where y = organ size, x = body size, b = allometric constant and a = allometric exponent.

In the present study the data obtained on oral and ventral suckers, pharynx and gonads in *G. tigrinum, G. korkei* and *G. punjabensis* were plotted against the body size and shown in Figs. 1–4. It is clear from these that sizes of similar organs of all species are falling in a linear line indicating the synonymity of *G. korkei* Bhalerao, 1936 and *G. punjabensis* Gupta, 1954 with *G. tigrinum* Mehra and Negi, 1928.



FIGS. 1–4. Fig. 1. Body size *vs.* oral sucker. Fig. 2. Body size *vs.* ventral sucker. Fig. 3. Body size *vs.* testes. Fig. 4. Body size *vs.* ovary. (· *G. tigrinum* ; *o*, *G. korkei* ; *x*, *G. punjabensis.*)

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---

1. Bhalerao, G. D., *J. Helm.*, 1936, 14, 181.
2. Gupta, N. K., *Res. Bull. Punj. Univ.*, 1954, 55, 125.
3. Klein, W., *Zool. Johrb.*, 1905, 22, 72.
4. Mehra, H. R. and Negi, P. S., *Alld. Univ. Studies,* 1928, 4, 63.
5. Narsimha Rao, L., *Curr. Sci.*, 1974, 43, 285.
6. Rohde, K., *Proc. Helm. Soc. Wash.*, 1966, 33, 184.

BIOLOGICAL CONTROL FOR THE PERNICIOUS WEED *PARTHENIUM HYSTEROPHORUS* LINN.

THE 'congress grass' *Parthenium hysterophorus,* whose seeds reached India with the wheat imported from Canada and U.S.A. has become naturalized in many parts of our country[1-4]. The weed is also harmful to human beings[5]. It causes eczema and affects lungs, eyes and nose[6]. The pollen grains of the weed floats in the air and cause several allergic diseases like dermatitis, fever and asthma[7]. Parthenin, one of the constituent principles characterized, is said to produce depressant effect on the nervous system[1]. Therefore it is high time that effective control measures are -launched for the eradication of this pernicious weed.

Several herbicides have been tried to check the growth of this weed[2]. Ansar 529 is a chemical based on arsenic that can prove fatal to human beings and cattle. The Indian tea board has banned the use of this chemical in tea gardens. Therefore it is not desirable to employ this in *Parthenium* control. The other herbicides, however, can cause less harm to animals, but these are expensive. In this context it may be of interest to note that a biological control has been successfully evolved to check this harmful weed by employing bugs of the species *Aphis fabae* (Fig. 1).

These hemipterans are well known for their quick fecundity ; the life cycle is completed in a few days and the females are parthenogenetic in most stages[8]. Kennedy and Booth[9] and Kennedy *et al.*[10] have already reported the remarkable ability of adaptation of these pests to new plant hosts.

These plant bugs are commonly present in *Ipomioea purga* growing in the environs of Coimbatore. When transferred to *Parthenium* they adapted well to the weed and produced desirable effects. Figure 2, presents photographs of two *Parthenium* plants grown under identical conditions in a green-house out of seeds planted at the same time. When they grew about 25 cm tall, the bugs were released on one of the plants. The infected plant grew only about 6 cm more in 4 months while the other plant reached 134 cm and started flowering profusely and in the normal way the fruits and seeds were also produced. In the infected plant, on the other hand, only a few flowers appeared and they never yielded fruits and seeds. This can be an ideal and most desirable effect. Experiments conducted in the field has also produced similar results. Further, since the viviparous females and males of the bugs are winged[8], they easily spread and cover larger areas within a short period.

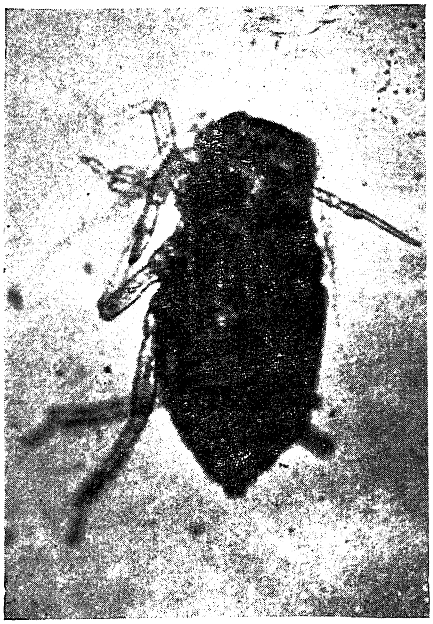

FIG. 1. A nymph of *Aphis fabae* which affects the weed more effectively than the adults.

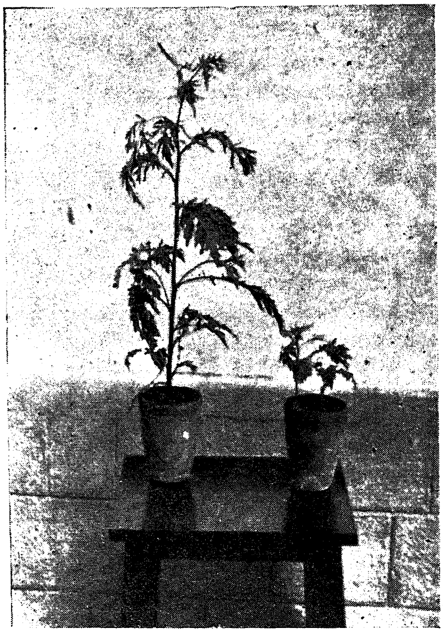

FIG. 2. *Parthenium hysterophorus* infected with *Aphis fabae* (right) and an uninfected plant (left).

Our grateful thanks are due to Professor G. R. Damodaran for his interest in this investigation.

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---

1. Chandras, G. S. and Vartak, V. D., *PANS*, 1970, 16, 212.
2. Jayachandra, A., *Curr. Sci.*, 1971, 40, 568.
3. Hakoo, M. L., *Ibid.*, 1963, 32, 273.
4. Maheswari, J. K., *Ibid.*, 1966, 36, 181.
5. Vartak. V. D., *Indian Fmg.*, 1968, 18, 23.
6. Maheswari, J. K., *Curr. Sci.*, 1968, 37, 326.
7. Anonymous, *News Letter from Agromore Ltd.*, Bangalore, 1970, 1 (5), 1.
8. Davidson, J., *Ann. appl. Biol.*, 1923, 10, 35.
9. Kennedy, J. S. and Booth, C. O., *Ibid.*, 1951, 38, 25.
10. —, Ibbotson, A. and Booth, C. O., *Ibid.*, 1950, 37, 651.

HISTOCHEMICAL LOCALIZATION OF NON-SPECIFIC ESTERASE ACTIVITY IN *PARAMPHISTOMUM CERVI* (TREMATODA : PARAMPHISTOMIDAE)

IN recent years much attention has been focussed on the histo-physiology of helminth parasites. Although studies have been made on various enzymes in digenetic trematodes, a few attempts have been made on the histochemical localization of esterase activity[1-6]. *Fasciola hepatica* has been studied extensively for carboxylic esterase activity[7-10]. The present work describes the histochemical localization of non-specific esterase activity and its probable significance in the digenetic trematode, *P. cervi*.

Live adult worms from the intestine of sheep were collected and washed in saline water for further processing. They were frozen rapidly with dry ice and sections were cut at 8 μ in Pearse–Slee Cyrostat (maintained at – 20° C). To detect the esterase activity the sections were incubated in the indoxyl acetate (0-acetyl-5-bromoindoxyl) medium[11] at 37° C for one hour. Sodium fluoride ($7 \cdot 5 \times 10^{-2}$ M) was used to inhibit the esterase activity.

The sites of the enzyme activity were marked by the deposition of dark blue granules in the tissues of the worm. The esterase activity in the oral sucker was moderate and the activity was restricted to the outer part of this organ revealing thereby that oral sucker has an important role in ingestion of host tissue as speculated by Becejac *et al.*[12]. Further, Allen and Reinard[6] attributed the secretory function of oral sucker to internal and/or extracarporeal digestion. The acetabulum exhibited weak to moderate reactions for esterase activity in contrast to the intense reaction reported by Becejac *et al.*[12] and Krvavica *et al.*[8]. However, esterase activity hitherto observed was associated with the outer lining of the acetabulum thereby indicated the attachmentary function as suggested in other trematodes[4,6]. Both cuticle and subcuticle exhibited moderate reaction for esterase activity which would indicate the secretion of the enzyme by the subcuticular cells and its further accumulation in the overlying cuticle. This process supports the suggestion made by Bogitsh[2] that the subtegumentary cells help to maintain the intimate contact with the host tissue by secretory functions.

The activity was more pronounced along the length of the caeca with slightly more intense reaction in the caecal contents (Fig. 1). Similar observations were made in other trematodes[7,13] but Ohman[13] reported the variations of esterase activity in the caecal cells of three different trematodes. The present observations clearly indicated that the caecum was more active for esterase activity than the cuticle which suggests that the caecum is more involved in the absorption and transfer of metabolites into the worm as reported in *Fasciola hepatica*[10]. The esterase activity in the parenchyma appears to be deposited in the form of small granules which were uniformly distributed throughout the tissue with slightly more granules accumulated in the cells surrounding the caeca. The parenchyma cells probably help in the supply of nutrients to the neighbouring tissues.

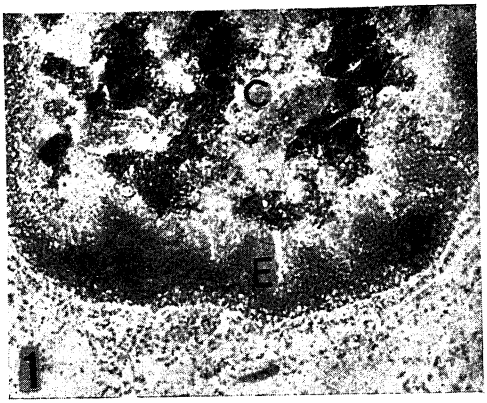

FIG. 1. Longitudinal section through part of the caecum showing intense esterase activity in the epithelial layer (E) and caecal contents (C), × 300.

The epithelial layer of testis and the cells of the ovary exhibited moderate reactions, while Allen and Reinard[6] and Halton[7] observed intense reaction in the epithelial layer of testis. Fully developed eggs within the uterus showed moderate activity. The vitellaria was found to be negative for this enzyme activity.

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---

1. Lee, D. L., *Parasitology,* 1962, **52**, 103.
2. Bogitsh, B. J., *Expt. Parasitology,* 1966 *a,* **19**, 339.
3. Fripp, P. J., *Ibid.,* 1967, **21**, 380.
4. Halton, D. W., *J. Parasitology,* 1968, **54**, 1124.
5. Howell, M. J., *Parasitology,* 1971, **62**, 133.
6. Allen, W. R. III and Reinard, H., *Proc. Helmith. Soc. Wash.,* 1972, **39**, 151.
7. Halton, D. W., *J. Parasitology,* 1967 *b,* **53**, 1210.
8. Krvavica, S. *et al., Expt. Parasitology,* 1967, **21**, 240.
9. Pantelouris, E. M., *J. Parasitology,* 1967, **53**, 354.
10. David, H. B., *et al., Expt. Parasitology,* 1968, **23**, 355.
11. Holt, S. J., (J. F. Danielli, ed.), *General Cytochemical Methods.* Acad. Press, New York, 1958.
12. Becejac, A. *et al., Veternarski Archiv.,* 1964, **34**, 87.
13. Ohman, C., *Parasitology,* 1966 *a,* **56**, 209.

THE CEREBRAL GLAND OF *GONOPLECTUS MALAYUS* (CARL.) (MYRIAPODA, DIPLOPODA)

THE neurosecretory system of invertebrates has been worked out in detail but very little work has been done in Myriapods. Very few workers have attempted to study the neurosecretory system of Chilopodes[1,2,4] and Diplopodes[3,5-8]. The present investigation on the neurosecretory system of millipede *G. malayus* has revealed the detailed structure of the cerebral gland.

The cerebral glands of the millipede under study are two in number, located in the lateral side of the brain as in *Jonespeltis*[6]. The glands are in close contact with the wall of a blood sinus and it is likely that the product of neurosecretion is discharged into this sinus. In living condition, the glands are bluish in colour. Each cerebral gland is connected to the brain by two nerves, the anterior and the posterior. The anterior nerve is 420 μ long and at the base its breadth is 140 μ while near the gland it is only 28 μ. The posterior nerve is longer and thinner in comparison to the anterior nerve. Its length is 700 μ and breadth only 28 μ (Fig. 1).

The shape and size of the cerebral gland vary according to the accumulation of neurosecretory product. Normally it measures about 490 μ in length but its breadth ranges from 160 μ to 170 μ whereas at the ends, its breadth is only 98 μ (Fig. 2). Unlike *Jonespeltis* (Prabhu, 1959) the boundary of the gland is smooth. Gabe[1,2] is of the opinion that the cerebral gland discharges its own secretion but in *G. malayus* it could not be confirmed. It appears to be a storage and releaser organ, and seems to resemble the sinus gland of crustacea.

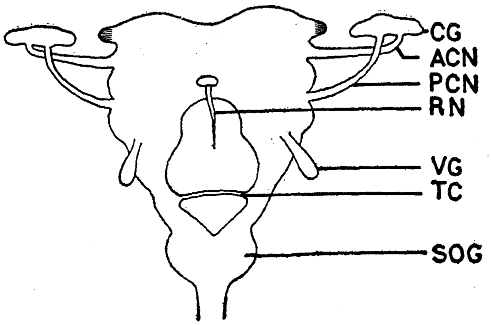

FIG. 1. Neurosecretory system of *Gonoplectus malayus* (Carl.).

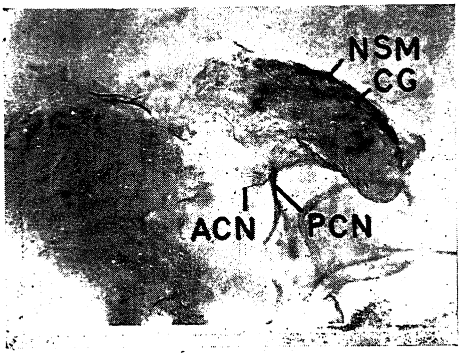

FIG. 2. Whole mount of cerebral gland associated with cerebral nerves and the part of the brain. P.F. *in situ*.

ACN, Anterior cerebral nerve; CG, Cerebral gland; NSM, Neurosecretory material; PCN, Posterior cerebral nerve; RN. Recurrent nerve; SOG, Suboesophagial ganglion; TC, Transverse; connective; VG, Visceral ganglion.

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---

1. Gabe, M., *C.R. Acad. Sci.* (Paris), 1952, 235, 1430.
2. —, *Bull. Soc. Zool.*, France, 1954 *a*, **78**, 338.
3. —, *C.R. Acad. Sci.* (Paris), 1954 *b*, **239**, 828.
4. Palm, N. B., *Ark. Zool.*, 1955, 9 (4), 115.
5. Prabhu, V. K. K., *Curr. Sci.*, 1959, **28**, 330.
6. —, *Z. Zellforsch.*, 1961, **54**, 717.
7. Sahli, F. and Petif, J., *C.R. Acad. Sci.* (Paris) (D), 1972, **275**, 2017.
8. —, *Symp. Zool. Soc.*, London, 1974, 23, 217.

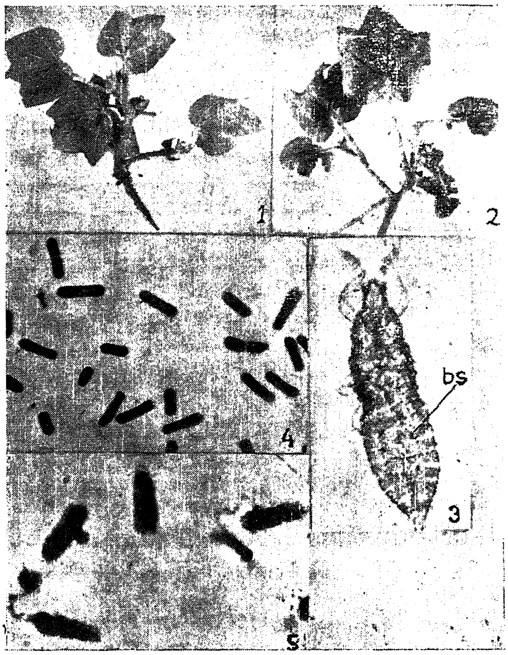
ON THE OCCURRENCE OF *BACILLIDIUM* SP. A MICROSPORIDIAN PARASITE INFECTING SOME THRIPS LARVAE

IN the course of investigations on the population dynamics of two Terebrantian thrips species—*Thrips flavus* (Schrank) and *Scirtothrips oligochaetus* (Karny), noted as pests of cotton, causing deformation and early drying of the leaves and resulting in the stunted growth of the plants, a dramatic decline in the population was observed during September 1975. An examination of available larvae and adults revealed the occurrence of the microsporidian parasite *Bacillidium* sp. and the infection was so intense, that in some larvae, all available space in the haemocoele was occupied by microsporidian spores. To our knowledge, this is the first record of any endoparasitic protozoan in a species of thrips.

The average number of thrips, adults and larvae on an infected plant, was about 110 larvae and 25 adults/10 cotton leaves. Later in the middle of October 1975, it was noted that the rate dropped approximately to 2 larvae and one single adult/10 cotton leaves. At the peak of infestation by the microsporidian, the infestation rate was nearly 100%, *i.e.*, almost all the adults and larvae suffered from the infection. Later thrips population almost completely disappeared at the end of October. The revival of the population was again observed at the end of December 1975. However, there was a slow rise in the population of thrips. The mild infection of *Bacillidium* sp. has been observed in the new population of thrips and the incidence of infection is on the increase. Detailed experimental studies on the mode of infection by the microsporidian and its life history are in progress.

The damage caused by *Thrips flavus* (Schrank) and *Scirtothrips oligochaetus* (Karny) is well known. *Thrips flavus* (Schrank) is a cosmopolitan polyphagous species, found on a great variety of plants. Morison (1957) collected it from 245 species of plants. Several records of the damage done by this species are by Van Ecke (1922), Priesner (1923), Bailey (1935), Speyer (1936), Morison (1943) and Ananthakrishnan (1969).

Bailey (1935) also noted that *Thrips flavus* (Schrank) is able to transmit mosaic virus of *Vicia faba.* The earlier record of *T. flavus* being a pest of cotton includes the work of Dyadechko (1964) (from the transcaucasian region of U.S.S.R.). Along with *Thrips florum* (Schmutz), this species has been considered a serious pest of tea flowers in India.



FIGS. 1–5. Fig. 1. Healthy cotton leaves. Fig. 2. Cotton leaves infested by *Scirtothrips oligochaetus* (Karny) and *Thrips flavus* (Schrank). Fig. 3. Second instar larva of *Thrips flavus* (Schrank) showing the *Bacillidium* spores (*bs*). Figs. 4–5. Smears showing different stages of *Bacillidium* sp.

Scirtothrips oligochaetus (Karny), found on several plants, was collected mainly from cotton leaves and flowers by the author. A closely related species or sibling species of *S. oligochaetus,* i.e., *S. dorsalis* has been recorded as a serious pest of chillies in South India (Ramakrishna Ayyar, 1932). The two species show many morphological similarities. It is of interest that the same microsporidian *Bacillidium* sp. can parasitise both the species of thrips and cause heavy mortality. Under natural conditions, this microsporidian may be responsible for bringing about fluctuations in population of thrips. Judging from the damage caused by these thrips to the economically important plants like cotton, it would be of great interest to use this microsporidian as a tool in the biological control of thrips. Earlier the biological control methods used in case of *Thrips tabaci* with chalcid parasite *Thripoctenus vuiletti* have been successsful.

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1. Ananthakrishnan, T. N., CSIR : *Zool. Monog.*, 1969, 1 (7), 118.
2. Bailey, S. F., *J. Econ. Ent.*, 1935, 28, 856.
3. Dyadechko, N. P., *Thysanoptera of the European Part of U.S.S.R.* (English Translation) Publ. Academic Council of Ukraine, Institute of Scientific Research for Plant Protection.
4. Morison, G. D., *Trans. R. ent. Soc.*, 1957, 23, 199.
5. —, *Ent. Mon. Mag.*, 1971, 106, 163.
6. Priesner, *H. Tijdschr. Ent.*, 1923, 66, 88.
7. Ramakrishna Ayyar, T. V., *Agric. Livstk. India,* 1932, 2, 391.
8. Sakimura, K., *Proc. Hawaii ent. Soc.*, 1947, 13, 59.
9. —, P.R.I. Tech. Paper No. 276, *Ann. ent. Soc. Am.*, 1960, 53, 1.
10. Speyer, E. R., *Rep. exp. Res. Sta. Chestnut.*, 1936, 21, 63.
11. Van-Eecke, R., *Naturk Verh. Holland Maatsch Wet.*, 1922, 9, 142.

A NEW WILT DISEASE OF MUSKMELON IN INDIA

MUSKMELON (*Cucumis melo* L.) is vulnerable to a number of fungal, bacterial and viral diseases[1]. Amongst fungal diseases, wilt caused by different species of Fusarium has been found to be responsible for serious loss to this crop. *F. solani* (Mart.) App. and Wellenw. was first found to be associated with the wilt of muskmelon in France[2]. Subsequently, the same pathogen has been reported from U.S.A.[3], Holland[4], Norway[5], Australia[6], Germany[7] and Israel[8]. From India, only *F. oxysporum* f. sp. *melonis* (Leach and Currence) Snyder and Hansen. has been isolated from wilted melon plants[9]. *F. solani* has been reported to cause fruit rot of Kakri (*Cucumis melo* var. *utilissimus* Duthie and Fuller) amongst the cucurbits from this country[10], but wilt of muskmelon caused by this pathogen is the first record from India.

The attack of Fusarium wilt of muskmelon was first detected in a number of samples from the Vegetable Research Farm of the Punjab Agricultural University, Ludhiana, in muskmelon growing season of 1972 and subsequently in the years 1973–75. The incidence of the disease is gradually increasing in the Punjab State, posing a serious threat to the cultivation of muskmelon in this State. Besides the University Research Farm, the pathogen has also been isolated from growers' muskmelon crop fields at Ludhiana, Patiala and Amritsar.

Significantly different types of symptoms are produced by different species of Fusarium attacking muskmelon plant[2,11-13]. By the pathogen isolated presently, the plants can be infected at any stage of growth but the symptoms are generally minifested when the plants are in flowering and bearing stage. The infected plants exhibit yellowing of foliage which later droop down due to the loss of turgidity. Soon the plants wilt completely, followed by the death of vines. The roots show vascular discoloration of orange red colour which extends to the stem to a small distance. The roots were otherwise perfectly healthy and normal without any sign of rotting. Stunting of vines was also exhibited to some extent. The pathogen causes yellowing of foliage and ultimate wilt but is not a rootrotter[14]. No masses of pink spores at the collar region were seen.

The causal organism was isolated on potato-dextrose-agar (PDA) and maintained on the same medium for further study. The pathogenicity was proved by inoculating the variety Hara Madhu of muskmelon. Pathogenicity tests were also carried on varieties, Lucknow Safeda, Pusa Sharbati and Arka Rajhasn and all these proved susceptible to the pathogen.

The fungus produced both micro- and macroconidia in culture. It was identified as *Fusarium solani* (Mart.) App. and Wellenw. (IMI Nos. 188227–188231) by the courtesy of Director of the Commonwealth Mycological Institute, Kew, London.

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Ludhiana, *December* 27, 1975.

---

1. Walker, J. C., *Diseases of Vegetable Crops*, McGraw-Hill Book Co., New York, 1952, p. 529.
2. Dufrenoy, J., *Ann. Epiphytics*, 1921, 7, 405.
3. Wiant, J. S., *Tech. Bull. U.S.D.A.*, 1937, No. 573, 47.
4. Var Koot, Y., *J. Tijdscher, Pl. Ziekt.*, 1943, 49, 52.
5. Roll–Hansen, F., *Meld. Norg. Landbr. Jojsk*, 1950, p. 257.
6. Conroy, R. J., *J. Aust. Inst. Agric. Sci.*, 1952, 19, 106.
7. Kosswig, W., *The Symptomatology of Fusarium Wilts of Cucumber* (C. sativus L.) *and Their Relation to the Wilt Disease of Other Plants*, Hamburg and Berlin, Paul Rarely, 1955, p. 148.
8. Palti, J. and Joffe, A. Z., *Phytopath. Z.*, 1971, 70, 31.
9. Bhaskaran, R., Prasad, N. M. and Kothandaraman, R., *Curr. Sci.*, 1971, 40, 171.
10. Suryanarayana, D., Chand, J. N., Kohli, C. K. and Raina, G. L., *J. Res. Punjab Agri. Univ.*, 1966, 3, 155.
11. Conroy, R. J., *Aust. J. Sci.*, 1961, 23, 412.
12. Toussan, T. A. and Snyder, W. C., *Phytopathology*, 1961, 51, 17.
13. Nikiforova, N. A., *Inst. Orosh. Z. Emled. Ovoshch.*. 1966, 5, 168.
14. Booth, C., *The Genus*, Fusarium, Commonwealth Mycological Institute, Kew, England, 1971, p. 237.

STIMULATION OF GERMINATION BY GAMMA RADIATIONS OF THE DORMANT SEEDS OF *AVENA FATUA* L.

SEEDS of *Avena fatua* L. (wild oats) are fully dormant at the time of harvest[1]. During dry storage the dormancy is gradually lost. Dormancy can also be overcome if imbibed seeds are given low temperature[2], gibberellic acid[3], high oxygen concentration[4], H_2O_2[4], KNO_3[4], etc., treatments. Germination inhibitors have been found in the hulls as well as the caryopses of *A. fatua* seeds[1,4]. Naylor and Simpson[3] attributed the dormancy in *A. fatua* seeds to a balance between germination promoters and inhibitors. It has been demonstrated that chilling and high oxygen concentration resulted in reduced levels of inhibitors[1,5].

Dormant seeds of *A. fatua* having 8% moisture content were exposed to 10, 20 and 30 kR Co^{60} gamma radiation at the dose rate of 900 r per minute and were immediately soaked in water, seeds were germinated in petriplates at 20° C. In seed lots where the maximum germination on 7th day was only 58%, better than 90% germination was obtained with 10 kR treatment (Fig. 1). Even in 30 kR treatment, stimulation of germination was observed but harmful effects of radiation became more pronounced, hence the germination rate was slower. Doses higher than 10 kR resulted also in retarded seedling growth (Fig. 2). Seedling growth was not depressed by 10 kR dose.

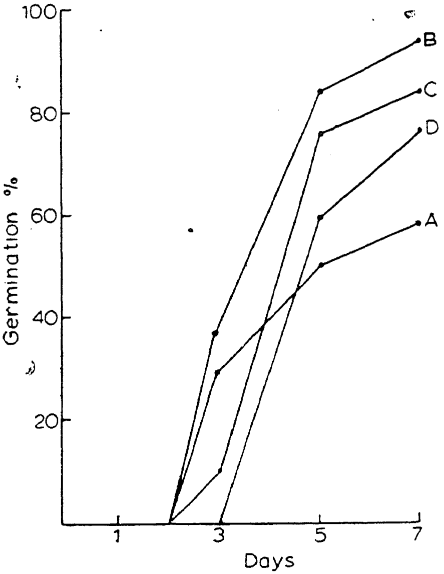

FIG. 1. Germination percentage of gamma-irradiated dormant *Avena fatua* seeds. A; control, B: 10 kR, C: 20 kR, D: 30 kR.

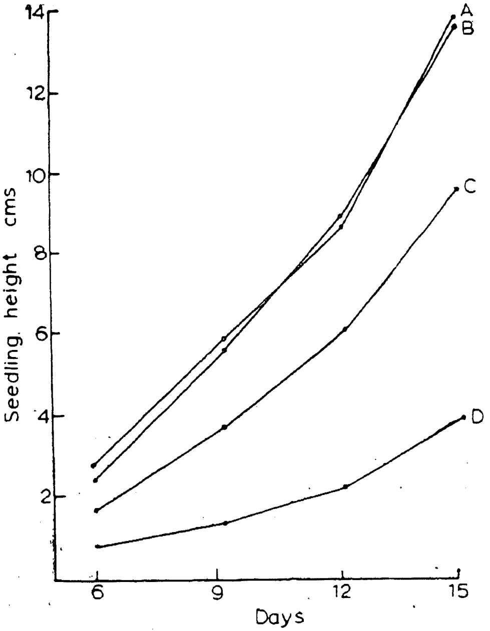

FIG. 2. Seedling height of *Avena fatua* seedlings raised from seeds exposed to gamma radiation. A: control, B: 10 kR, C: 20 kR, D: 30 kR.

Considering the evidence that gamma radiations cause the production of H_2O_2 in the tissues[6] and increased oxygen uptake and respiration[7,8] along with the fact that treating the seeds with H_2O_2[4] and placing the seeds in high oxygen concentration[1] promote germination of dormant seeds, it seems quite likely that gamma radiations promote germination of dormant *A. fatua* seeds by increasing their oxygen uptake and causing formation of peroxy radicals in the tissues, which in turn may inactivate the inhibitors by oxidation and thus tilting the promoter/inhibitor balance in favour of the promoters. It was suggested by Sax[9] that ionizing radiations may stimulate plant growth by altering the auxin balance in the organism, but only a few studies[10,11] have been conducted on the effects of ionizing radiations on different plant growth regulators. Gamma-irradiated dormant *A. fatua* seeds thus provide an excellent system in which the mechanism of stimulation of plant growth by radiation can be studied.

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---

1. Black, M., *Can. J. Bot.*, 1959, 37, 393.
2. Friesen, G. and Shebeski, L. H., *Weeds*, 1961, 9, 634.
3. Naylor, J. M. and Simpson, G. M., *Can. J. Bot.*, 1961, 39, 281.
4. Hay, J. R., *Ibid.*, 1962, 40, 191.
5. Wareing, P. F. and Foda, H. A., *Physiol. Plant*, 1957, 10, 266.
6. Casarett, A. P., *Radiation Biology*, Prentice Hall, Inc., 1968.
7. Woodstock, L. W. and Justice, O. L., *Radiat. Bot.*, 1967, 7, 129.
8. Sidrak, G. H. and Sucss, A., *Ibid.*, 1973, 13, 309.
9. Sax, K., *Ibid.*, 1963, 3, 179.
10. Gordon, S. A. and Weber, R. P., *Plant Physiol.*, 1955, 30, 200.
11. Sideris, E. G., Nawar, M. M. and Nilan, R. A., *Radiat. Bot.*, 1971, 11, 209.

BLOSSOM END ROT OF BOTTLEGOURD (*LAGENARIA SICERARIA*) (MOLINA) STANDL. INCITED BY *PYTHIUM BUTLERI* SUBR.

A VERY high incidence of blossom end rot on bottlegourd fruits was noticed at Hessaraghatta farm in trials against powdery mildew with systemic fungicides, during January–February, 1975. Fruits of all ages showed the attack though the incidence was more on young developing ones.

Infection started from blossom end as small water soaked lesions which gradually turned darkish and progressed towards stem end covering the entire tip region. Under humid conditions the diseased tissues were covered by a whitish, fluffy, mycelial growth. In certain cases the entire fruit was profusely covered. The flesh turned brownish and became corky.

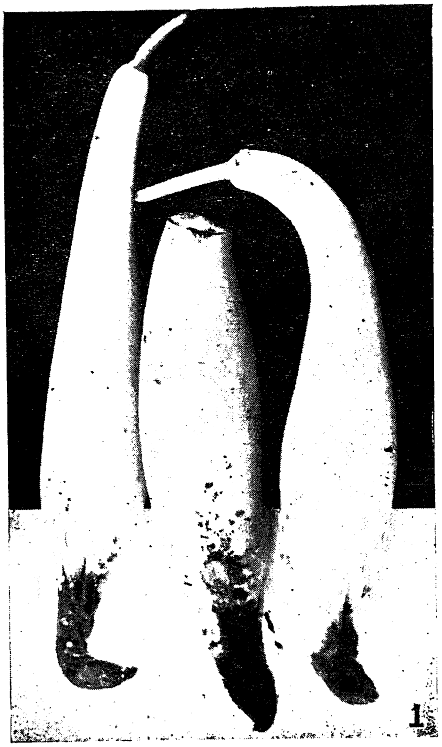

FIG. 1. Typical field symptoms of blossom end rot on bottlegourd fruits.

Pythium butleri Subr. was isolated from the infected fruits in pure culture and produced typical disease symptoms on artificial inoculation within 36 hours. Fruits of watermelon, muskmelon, pumpkin, cucumber, coccinia, roundmelon and squash (patty pan) were inoculated. All were found susceptible. The fungus is known to attack various hosts[1-6], but so far it has not been reported to cause fruit rot of cucurbits. The fungus has been deposited under I.M.I. No. 167978.

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---

1. Drechsler, C., *Sydowia*, 1955, 9, 451.
2. Srivastava, D. N. and Rao, V. R., *Curr. Sci.*, 1964, 33, 119.
3. Aulakh, K. S., *Indian Phytopath.*, 1971, 24, 611.
4. Reddy, H. R. and Anil Kumar, T. B., *Curr. Sci.*, 1975, 44, 407.
5. Singh, S. L. and Pavgi, H. S., *Ibid.*, 1975, 44, 487.
6. Sokhi, S. S. and Sohi, H. S., *Ind. Jour. Hort.*, 1975, 32, 182.

FUNGI IN THE ROOT REGION OF *PARTHENIUM HYSTEROPHORUS* LINN.

Parthenium hysterophorus L., an allergic weed, belonging to the family Compositae has been reported to contain inhibitors in all parts of the weed[1]. Further, it is well-known that the seat of synthesis and the storage of alkaloids are the young root tip and the root bark, respectively. It seemed, therefore, probable that these alkaloids might have a significant influence on the microbial population on and around the roots. Hence the rhizosphere and rhizoplane mycoflora of *P. hysterophorus* were examined.

Plants were collected at the pre- and post-flowering stages at random around the Central College Campus, Bangalore. Roots, with the rhizosphere soil still attached were carefully removed. The control soil was collected 10–12 cm away from the root. Samples were stored in polythene bags and processed in the laboratory. The soil from the roots was removed by tapping in measured quantity of sterile water. Dilutions were made from the suspension. Waksman's dilution plate technique was followed for the isolation of the fungi[2,3]. One ml. of 1 : 10,000 dilution was plated on martin rose-bengal agar using streptomycin as antibacterial agent[4]. For the rhizoplane mycoflora, the roots cleared of the rhizosphere soil were cut into 1·0 cm pieces and plated on the same medium.

Results

A general suppression of fungal species in the pre-flowering and the post-flowering rhizosphere soil is noticed (Table I). It seems probable that the presence of alkaloids in the root region is correlated with the suppression of fungi[5]. The increase in the fungal population in the rhizosphere soil of post-flowering plant could be explained as due to a change in the composition of root exudates or it may be due to more root leakage at the time of the maximum vegetative growth, and least in the post-flowering stage[6]. Further, Tables I and II show that there is complete suppression of *Penicillium* species both in the rhizosphere (pre- and post-flowering) and on the rhizoplane of *P. hysterophorus*,

TABLE I

Fungal species from the rhizosphere and control soil of Parthenium hysterophorus

Fungal species	Pre-flowering stage	Post-flowering stage	Control
Alternaria tenuis	−	+	−
Aspergillus flavus	+	+	+
A. niger	+	+	+
A. ochreous	+	+	+
A. sulphureus	+	+	−
A. candidus	+	+	+
A. nidulans	+	+	−
Acremonium vitis	−	+	−
Acrostalagmus sp.	−	+	−
Circinella simplex	−	+	−
Cladosporium herbarum	+	+	+
Cunninghamella echinulata	+	−	−
Curvularia lunata	+	+	−
Fusarium sp.	+	+	+
Helminthosporium tetramera	−	+	−
Mucor racemosus	+	+	+
Mortierella sp.	−	+	−
Myrothecium roridum	−	+	−
Penicillium sp.	−	−	+
Phoma glomerata	−	+	−
Pythium intermedium	−	+	−
Rhizopus nigricans	+	+	+
Starkeomyces Koorchala moides	−	+	−
Theilavia terricola	−	+	−
Trichoderma viridis	−	+	−
Verticillium alboatrum	−	+	−
Paecilomyces sp.	+	+	−

TABLE II

Fungal species on the rhizoplane of Parthenium hysterophorus

Fungal species	Pre-flowering root bits	Post-flowering root-bits
Aspergillus niger	+	+
Cladosporium herbarum	+	+
Curvularia sp.	−	+
Fusarium sp.	−	+
Neocosmopara vasinfectum	−	+
Pyrenochaeta sp.	−	+
Rhizopus nigricans	+	−

but species of *Aspergillus* and *Cladosporium* could be isolated from all the platings. This clearly indicates that the root exudates can influence the composition of the root microflora[7]. Further work is in progress.

The author is grateful to Professor M. Nagaraj for providing the necessary facilities.

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1. Sukhada, D. Kanchan, *Curr. Sci.*, 1975, **44**, 10, 358.
2. Waksman, S. A. and Fred, E. B., *Soil. Sci.*, 1922, **14**, 27.
3. Warcup, J. H., In : *Ecology of Soil Fungi*, Liverpool Univ. Press, Liverpool, 1960, p. 3.
4. Martin, J. P., *Soil. Sci.*, 1950, **69**, 215.
5. Padma, R. and Mukherjee, K. G., *Indian Phytopath.*, 1972, **25** (1), 104.
6. Srivastava, V. B., *Ph.D. Thesis*, Univ. Gorakhpur, 1969.
7. Katznelson, H., In : *Ecology of Soil Borne Plant Pathogens*, Univ. of California Press, Berkeley, Los Angeles, 1965.

SIGNIFICANCE OF NITRATE REDUCTASE ACTIVITY IN DEVELOPING COTTON SEEDS

NITRATE reductase (NR) is the key regulator of influx[1] of reduced nitrogen, since it is the first enzyme involved in the biosynthesis of amino-acids. The activity of nitrate reductase is mainly associated with the photosynthetic[2] cells. However, light can regulate its activity in the organ[3], situated at a distance from the green tissues *via* the supply of sugars, energy and reducing factors. There occurs a sudden upsurge[4] in the levels of soluble nitrogen and proteins in the immature cotton seeds, just after fertilization. It is obscure[5] whether the activity of nitrate reductase in the developing seeds supports the synthesis of amino acids and proteins in the seeds.

Gossypium arboreum L. var. G 27, *G. hirsutum* L. var. J 34, J 205, LSS, H 297, RS 1, RS 8, RS 11 and RS 89 were grown in the field in the *Kharif* 1974, following all the package of practices recommended by Punjab Agricultural University. Ovules were sampled from selfed flowers at pre-fertilization, fertilization and post-fertilization stages for the determination[6] of the nitrate reductase activity.

Unfertilized ovules from all the varieties showed nitrate reductase activity. There was observed a fertilization-induced increase in the enzymic activity, regardless of the variety. Maximum activity was obtained in the one-day old fertilized ovules. A gradual decrease in the activity, thereafter, continued until 32-day old seeds. It appeared that there was no significant genotypic variation, rather it was the developmental stage of the seed which made the difference in the NR activity in the seed. Thus, a common type of change in ten varieties is being used here to generalize the process in cotton as whole (Fig. 1).

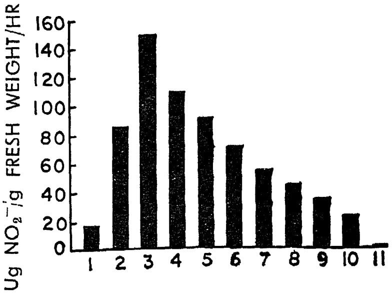

FIG. 1. Nitrate reductase activity in developing Cotton Ovules/Seeds. 1—Anthesis; 2—Fertilization; 3—One-day after fertilization; 4—Four-day after fertilization; 5—Eight-day after fertilization - - - - - - -; 11—Thirty-two-day after fertilization.

Nitrate reductase is mainly a substrate[1] (No_3^-) inducible enzyme. However, plant growth regulators[7] also mediate the enzymic activity. In the case of cotton, fertilizations proceeds the synthesis[8] of auxins, gibberellins and cytokinins, which support the development of seed as well as cellulose fibre. During the early phase of seed development, protein synthesis is intense and nitrogen to be utilized in protein formation in the seeds is unlikely to be transported as nitrate ions, but in the form of organic nitrogen as amides and amino-acids (Woodruff[9], 1972). However, a positive relationship between enzymic activity and seed weight suggests that nitrate reductase activity in the seed is, somehow, associated with the seed development in cotton.

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Ludhiana, *December* 29, 1975.

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1. Ellrich, G. L. and Hageman, R. H., *Crop. Sci.*, 1973, 13, 59.
2. Trawis. R. L., Huffakar. R. C. and Key, J. L., *Pl. Physiol.*, 1970, 46, 800.
3. Wallace. W., *Ibid.*, 1973, 52, 197.
4. Singh, O., *Ph.D. Thesis*, Punjab Agric. University, Ludhiana, 1975.
5. Nair. T. V. R. and Abrol, Y. P., *Experientia*, 1974, 28, 1480.
6. Joworski, E., *Biochem. Biophys. Res. Commun.*, 1971, 43, 1274.
7. Sharma. S. K., *M.Sc., Thesis,* Punjab Agric. University, Ludhiana, 1974.
8. Singh, O. and Singh, O. S., *Indian J. Expt. Biol.*, 1975, 13, 4111.
9. Woodruff, D. R., *Expt. Agric. Animal Hub.*, 1972, 12, 511.

FUNGITOXICITY OF SOME UREA DERIVATIVES OF 1, 3, 4-THIADIAZOLE

RECENTLY some mercapto and acetoxy derivatives of 1, 3, 4-thiadiazole have been reported to possess strong antifungal activity[1,2]. This prompted the authors to undertake the investigation of urea derivatives of 1, 3, 4-thiadiazole for their fungi toxicity and to evaluate their efficacy against standard fungicides.

The fungi toxicity of the compounds and commercial fungicides was observed against *Pythium aphanidermatum* by the poisoned food technique[3]. Requisite quantities of all the compounds were dissolved in a solution of acetone and water (5 : 95) so as to give a concentration of 5,000, 10,000, 20,000, 30,000 and 40,000 μg/ml. The same concentrations were maintained for the commercial fungicides in sterilized double distilled water. One ml of the prepared solution was added to presterilized petriplates containing nine ml of sterilized Czapek's agar medium so as to make a final dose of 500, 1,000, 2,000, 3,000 and 4,000 μg/ml of the compounds or fungicides in the medium. For control nine ml of the medium was supplemented with one ml of acetone : water (5 : 95) solution. A mycelial disc of 5 mm in diameter from the periphery of a 7-day old culture of the test fungus was inoculated upside downwards to assay plates. The plates were incubated at 28° C (± 1) and observations were recorded on the seventh day. The colony diameter of the test fungus was measured in mutually perpendicular directions and % mycelial inhibition calculated. Fungi toxicity of the compounds as well as of fungicides were measured in terms of toxicity index[4]. Experiments were repeated twice and each contained five replicates.

Out of seven urea derivatives of 1, 3, 4-thiadiazole, only two exhibited maximum toxicity index indicating their strong fungi toxicity at a minimum dose of 500 μg/ml (Table I). The toxic doses of both the compounds (4 and 5 in Table I) were four times less than that of commercial fungicides, *viz.*, Dithane Z–78 and Dithane M–45 while six times less than Ziram, indicating thereby that the compounds may be utilised as a new, more potent, fungi toxic agent on commercial basis. Table I also showed that the change in the position of nitrophenyl group of both the compounds (4 and 5 in Table I) had no effect on their fungi toxicity. Further studies to determine *In vivo* efficacy of both the compounds are in progress.

TABLE I

Comparative fungi toxicity of urea derivatives of 1, 3, 4-thiadiazole with commercial fungicides against Pythium aphanidermatum

Sl. No.	Name of compounds and fungicides	Toxicity index	Lethal dose in μg/ml
	*Compounds**		
1.	1-[(5-Phenyl-1, 3, 4-thiadiazole-2-yl)-3-(4-methyl)] benzene sulphonyl urea	339	2000
2.	1-[5-(4′-Flurophenyl-1, 3, 4-thiadiazole-2-yl)-3-(4-methyl)] benzene sulphonyl urea	252	3000
3.	Benzyl-[5-(4′-flurophenyl amino)1, 3, 4-thiadiazole-2-yl] sulphonyl urea	32	<4000
4.	1-[5-(3′Nitrophenyl-1, 3, 4-thiadiazole-2-yl)-3-(4′phenyl) thiazole-2-yl] urea	500	500
5.	1-[5-(4′-Nitrophenyl-1, 3, 4-thiadiazole-2-yl)-3-(4′-phenyl) thiazole-2-yl] urea	500	500
6.	1-[5-(4′-Nitrophenyl-1, 3, 4-thiadiazole-2-yl)-3-(5′methyl) thiazole-2-yl] urea	402	2000
7.	1-[5-(4′-Nitrophenyl-1, 3, 4-thiadiazole-2-yl)-3-(4′-methyl)] benzene sulphonyl urea	252	3000
	Commercial fungicides		
8.	Dithane M–45**	341	2000
9.	Dithane Z–78***	333	2000
10.	Ziram*	309	3000

* 100% active ingredient.
** 75% active ingredient.
*** 80% active ingredient.

Authors are thankful to Prof. K. S. Bhargava for necessary laboratory facilities; to Dr. S. N. Dixit for encouragement and guidance and to S.C.S.I.R. (U.P.) for financial assistance.

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Gorakhpur, *February* 17, 1976.

---

1. Srivastava, C. M., Dixit, S. N. and Rao, R. P., *Proc. Nat. Acad. Sci. 44th Session*, 1974, 2, 73.
2. —, — and Singh, H., *Proc. Symp. Plant. Disease Problem, Udaipur*, 1975, p. 43.
3. Grover, R. K. and Moore, J. D., *Phytopath.*, 1962, 52, 876.
4. Rich, S. and Horsfall, J. C., *Bull. Commecitant Agric. Exp. Sta.*, New Havan, 1961, p. 639.

A CONVENIENT NEW MEDIUM FOR RAPID PREPARATIONS OF CUTICULAR REPLICAS

CUTICULAR replicas are of value in taxonomic studies where surface details of the leaf epidermis are taken into consideration. Many different substances are utilised for obtaining such replicas including the mucilage from unripe fruits of *Coccinia cordifolia* Cong. (Patel, 1968)[1]. Sinclair and Dunn (1961)[2] and Dilcher (1974)[3] list the various media that have been used by different research workers for this purpose. The author noticed that the insulating material commercially branded 'Thermocole' dissolves rapidly in xylene giving a clear viscous fluid that can successfully be employed as yet another medium for obtaining cuticular replicas quickly.

Small pieces of clean Thermocole when dropped into a vial dissolve rapidly in dry xylene at room temperature. The air bubbles that appear when the Thermocole goes into solution may be removed by heating the solution to about 50° C for a few minutes. The solution thus obtained is clear and viscous, lending itself to be applied with a camel hair brush on to the surface of the leaf whose replica is desired. The solution spreads itself evenly on the surface and within 10 to 15 minutes dries up completely forming a very thin, slightly opaque film that can easily be peeled off either by hand or with tweezers. A fully dried film is quite firm showing no elasticity and can either be mounted dry or mounted in a drop of water for examination under transmitted light. A 10% solution was found to be a satisfactory medium for obtaining thin films of replicas from glabrous leaves such as that of *Bougainvillaea glabra* Choisy (Fig. 1). Since peels of fairly large dimensions can be obtained, they may be stored in small envelopes for future use,

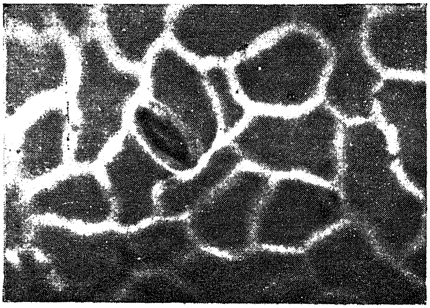

FIG. 1. Photomicrograph of the cuticular replica from the leaf of *Bougainvillaea glabra* Choisy, × 320.

This new medium may also be used for obtaining cuticular transfers from fossil compressions.

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1. Patel, R. J., *Curr. Sci.*, 1968, 37, 680.
2. Sinclair, C. B. and Dunn, D. B., *Stain Tech.*, 1961, 36, 299.
3. Dilcher, D. L., *Bot. Rev.*, 1974, 40, 120.

A NEW VIRUS DISEASE OF ARHAR [*CAJANUS CAJAN* (L.) MILLSP.]

Cajanus cajan (L.) Millsp. commonly known as "arhar" or pigeon pea is an important pulse crop of India. Recently, some plants of arhar showed symptoms quite distinct from the earlier ones[1-6]. Such symptoms were of two types, in the first type the diseased plants showed mild mosaic mottling and slight distortion and reduction in size of leaflets (Fig. 1). Also the affected plants were dwarfed and bore smaller fruits. Virus isolate, obtained from this plant, is referred to as AMM in the text. The second type of symptom consisted of irregularly scattered chlorotic and dark areas of varying shape and size. The leaflets were twisted and reduced in size (Fig. 2). Plants were dwarfed and bore smaller and fewer pods. Virus isolate obtained from such a type of plant is referred to as ASM in the text. The cross-protection tests, indicated that AMM and ASM were related strains of a single virus. The paper reports this new sap transmissible virus causing mosaic diseases in arhar from India.

FIG. 1. Leaves of arhar *cv.* Sarda infected with AMM isolate showing vein-clearing, mild mosaic and reduction of leaflets.

Several plants were inoculated separately by rubbing the leaves with the sap from the diseased plants of AMM and ASM. Both the isolates were similar in their host-range. They systemically infected *Arachis hypogaea* L., *C. cajan*, *Cassia occidentalis* L., *C. tora* L., *Cicer arietinum* L., *Clitoria ternatea* L., *Crotalaria juncea* L., *C. striata* DC., *Dolichos biflorus* L., *Indigofera hirsuta* L., *Lathyrus odoratus* L., *L. sativus* L., *Lens esculenta* Moench, *Lupinus albus* L., *Medicago denticulata* Willd.. *Melilotus alba* Desr., *M. indica* All., *M. parviflora* Desf., *Phaseolus aureus* Roxb., *P. calcaratus* Roxb., *P. mungo* L., *Pisum sativum* L., *Tephrosia purpurea* (L.) Pers., *Trifolium alexandrinum* L., *T. corniculata* L., *T. pratense* L., *Trigonella foenum-graecum* and *Vigna sinensis* Savi. Isolate AMM was mild in its reaction to the host plant as compared with ASM.

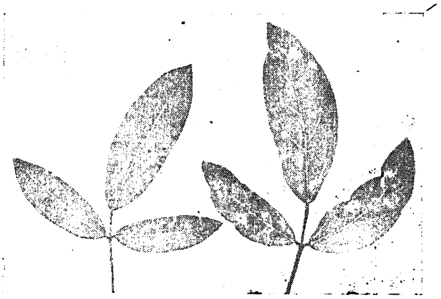

FIG. 2. Leaves of arhar *cv.* Sarda infected with ASM isolate showing dark green blotches which interspersed with chlorotic areas and reduction of leaflets.

The physical properties of the virus *in vitro* are given in Table I.

TABLE I
Physical properties of the virus isolates

Physical properties	Virus isolates	
	AMM	ASM
Tolerance of dilution	Between 10^{-5} and 10^{-6}	Between 10^{-5} and 10^{-6}
Resistance to heat	60°–65° C	80°–85° C
Longevity *in vitro*	16–17 days (22°–25° C)	11–12 days (22°–25° C)

The virus isolates retained infectivity in arhar *cv.* Sarda leaves desiccated over calcium chloride, for more than 735 days at 15°–31° C. The virus particles can remain active for longer duration under desiccation, due to cessation of several interacting factors which could retard the longevity of the virus, *e.g.*, pH of the extract and changes in pH with time and action of some enzymes like proteases or nucleases that may be initially present in the extract or released later by cell debris or micro-organisms containing the extract[7].

Virus isolates were easily transmitted by sap but could not be transmitted through the seeds or by any of the six aphids tested, *viz.*, *Aphis craccivora* Koch., *A. gossypii* Glove., *A. nerii* Boyer, *Lipaphis pseudobrassicae* Davis, *Myzus persicae* Sulz. and *Rhopalosiphum maidis* Fitch.

The infective sap showed negative reactions with the antisera against yellow mosaic virus, common bean mosaic virus, clover yellow mosaic virus, cowpea mosaic virus, potato virus X, red clover vein mosaic virus, southern bean mosaic virus, white clover virus and Wisconsin pea mosaic virus.

The symptoms produced on different hosts and physical properties of the virus under investigation are not in general agreement with those of other viruses infecting arhar, *viz.*, cowpea mosaic virus[2,9], yellow and pale mosaic virus[5] and common bean mosaic virus[8]. Also the present virus differs in its host-range, reaction and physical properties with the legume viruses limited to leguminosae[10]. The study indicates that the virus under discussion is an undescribed one. Therefore, it is proposed to designate it by a specific name as "arhar (pigeon pea) mosaic virus".

We are thankful to Prof. K. S. Bhargava, for facilities, Prof. C. W. Kuhn, Dr. M. Z. Maat and Dr. Koening for the supply of antisera.

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Gorakhpur, U.P., *August* 21, 1975.

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1. Su, V., *Intern. Bull. Plant Prot.*, 1931, 5, 1441.
2. Dale, W. T., *Ann. appl. Biol.*, 1949, 36, 327.
3. Capoor, S. P., *Indian J. agric. Sci.*, 1952, 22, 271.
4. Bisht, N. S. and Banerjee, A. K., *Labdev. J. Sci. Technol.*, 1965, 3, 271.
5. Newton, W. and Peiris, J. W. L., *F.A.O. Pl. Bot. Bull.*, 1953, 2, 27.
6. Williams, F. J., Grewal, J. S. and Amin, K. S., *Pl. Dis. Reptr.*, 1968, 52, 300.
7. Matthews, R. E. F., *Plant Virology*, Acad. Press, London and N.Y., 1970, p. 441.
8. Zaumeyer, W. J. and Goth, R. W., *Phytopath.*, 1964, 54, 1378.
9. Adsuar, J., *J. Agric. Univ. Puerto Rico*, 1964, 48, 264.
10. Bos, L., *Neth. J. Plant Path.*, 1964, 70, 161.

EFFECT OF LECANORIC ACID ON MITOTIC DIVISION IN *ALLIUM SATIVUM*

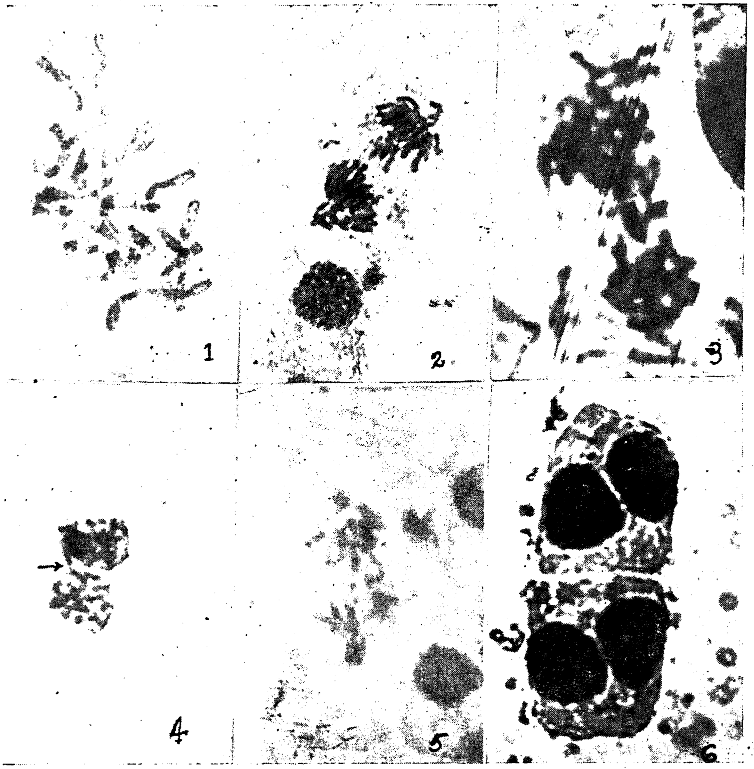
THE phenolic groups of chemicals have been reported to have mutagenic action on dividing chromosomes and inducing fragmentation[4-9]. Levan and Tjio[5,6] working with a number of phenols including pyrogallol, guaiacol, hydroquinone and resorcinol reported a high degree of chromosomal aberrations due to the effect of phenolic group. The effect of lecanoric acid, on dividing cells, has been presented in this note.

The actively growing root tips of *Allium sativum* were dipped in 0·1 M aqueous solution of the acid for 1, 2 and 3 h. at room temperature, between 10 A.M. and 1 P.M. The treated root tips were clipped and fixed in 1 : 3 acetic alcohol and kept in the refrigerator for 24 hours, and then transferred to 70% alcohol for preservation. The root tips were squashed in aceto orcein.

In general the chromosome condensation was delayed and was not complete even by metaphase (Fig. 1). The spindle was formed diagonally in the cell (Fig. 2). Some lagging chromosomes were observed at the equator while the chromatids of other chromosomes already reached the poles (Fig. 3). The entire chromosomes were seen connecting two sister nuclei (Fig. 4). This is probably due to the extreme delay in the division of centromeres of these chromosomes. Split spindles too were observed (Fig. 5) due to which chromosome groups of different sizes were seen scattered.

The appearance of diagonally arranged unequal sister nuclei in a cell were of common occurrence (Fig. 6). The double nucleated, cell formation is attributed to the failure of cell plate formation, during mitosis under the effect of the chemical.

The formation of diagonal spindle and inhibition of cell plate formation are reported earlier by several workers[2]. The phenomena are reported to be common with the action of methylated and alkylated oxypurines, such as caffeine, theophylline and theobromine on dividing cells. The inhibition of cell plate formation naturally results in the appearance of binucleated cells. The appearance of unequal sister nuclei is of some interest. Kihlman[2] attributes such a phenomenon to the fusion of nuclei in unequal number in a multinucleated cell. In the present preparations multipolar or split spindles were seen even though multi-nucleated condition was not observed. Probably the fusion of these chromosomal groups in unequal number might have given rise to two unequal sister nuclei in a cell. In addition, the inclusion of lagging chromosomes into one of the two sister nuclei might have further added to its size. The appearance of the lagging chromosomes was noted earlier by several workers[2,4]. They attribute this phenomenon to the non-synchronised division of the centromeres. The presence of an entire chromosome, connecting the two daughter nuclei (Fig. 4), seems to strengthen this presumption. High fragmentation of chromosomes through the action of phenols is reported earlier by several workers[5,6,8,9], yet no significant fragmentation could be observed in the present experiment for 1–3 hrs of treatment with 0·1 M solution of the acid.



FIGS. 1–6. Fig. 1. Non-condensed chromosomes at metaphase. Fig. 2. Diagonally arranged spindle with lagging chromosomes. Fig. 3. Telophase with diagonally arranged unequal sister nuclei and lagging chromosomes at the equator. Fig. 4. Arrow shows a chromosome with two chromatids in each arm and connecting sister nuclei. Fig. 5. Split spindle with groups of chromosomes of unequal sizes. Fig. 6. Double nucleated condition with diagonally arranged sister nuclei.

On the whole lecanoric acid seems to imbibe in it, the effects of methylated and alkylated oxypurines in inducing the appearance of diagonal spindle and unequal sister uuclei in a single cell.

We are highly thankful to Dr. P. S. Rao, Reader in Chemistry, Post-Graduate Centre, Vidyaranyapuri 506 009, Warangal, for providing us the chemical.

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1. Kihlman, B., *Hereditas*, 1949, 35, 393.
2. —, *Action of Chemicals on Dividing Cells*, Prentice Hall, Inc., Englewood, Cliff, New Castle, 1966.
3. Mehra, P. N. and Mann, S. K., *The Nucleus*, 1974, 17 (3), 167.
4. Chatterjee, P. and Sharma, A. K., *Ibid.*, 1972, 15 (3), 214.
5. Levan, A. and Tjio, J. H., *Hereditas*, 1948 *a*, 34, 250.
6. Levan, A. and Tjio, J. H., *Ibid.*, 1948 *b*, 34, 453.
7. Sharma, A. K. and Bhattacharya, N. K., *Genetica.*, 1956, 28, 121.
8. — and Choudhury, R. K., *Biologia*, 1963, 18, 126.
9. — and Sharma, A., *Int. Rev. of Cytol.*, 1960, 10, 101.

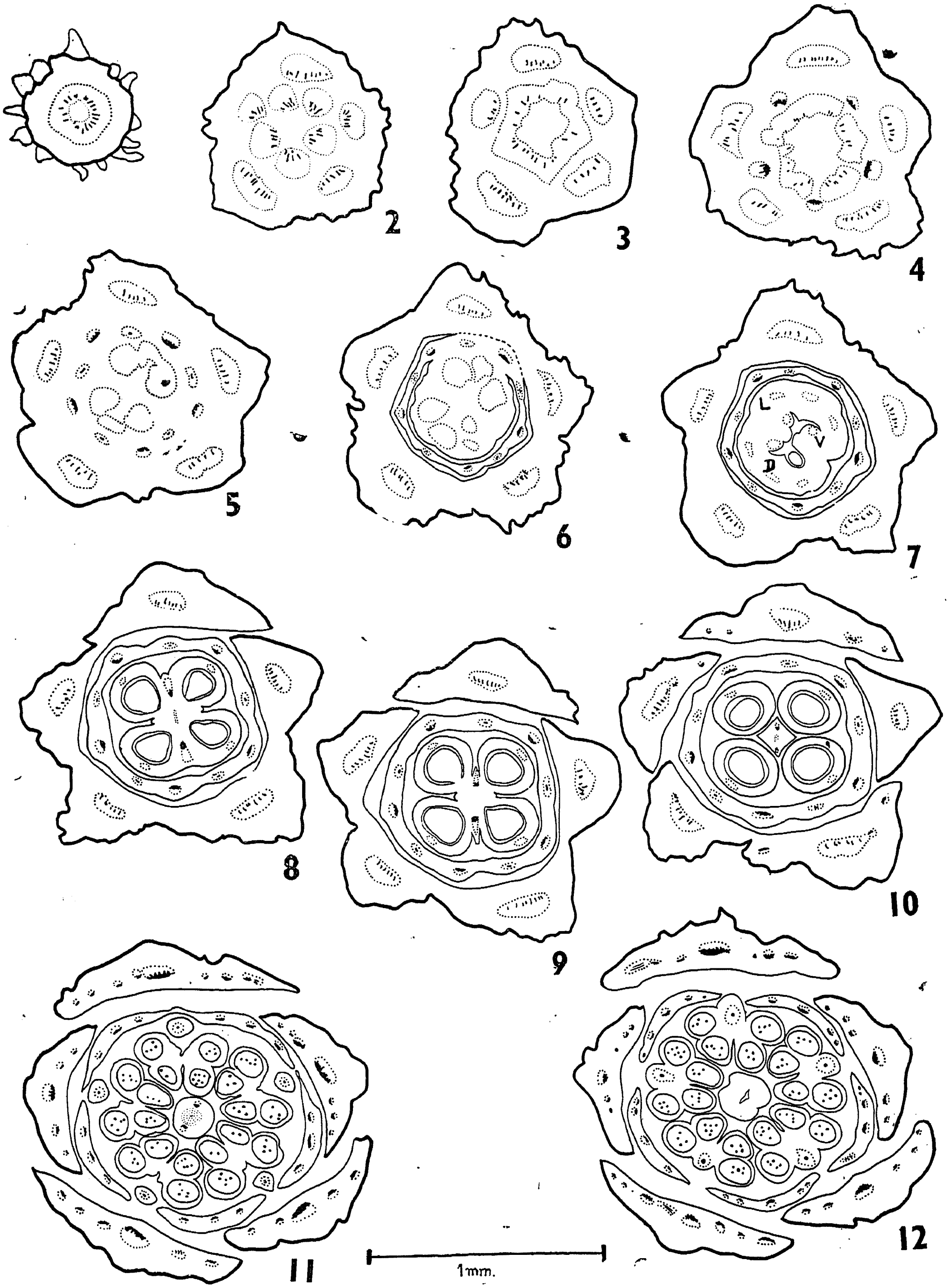
FLORAL ANATOMY OF BORAGINACEAE

I. *Sericostoma pauciflcrum* Stocks

THE family Boraginaceae has earlier attracted the attention of those interested in floral anatomy. Joshi[1], Lawrence[2] and Sharma[3] have described the anatomy of the flower in several genera but *Sericostoma* remained untouched. As it shows certain points of interest the floral anatomy of *S. pauciflorum* Stocks has been described in the present paper. The material was collected from

Okha and Dwaraka and usual techniques for fixation, cutting and staining have been employed.

The pedicel has a ring of vascular tissue which gives out five calyx traces at the base of the receptacle (Figs. 1–3). All the calyx traces get arranged at the periphery of the receptacle which is later separated as calyx tube (Figs. 4–7). Calyx splits up at the top into five quincuncially arranged



FIGS. 1–12. Serial transverse sections of the flower from base upwards showing the course of vascular supply. (D, Carpellary dorsal; L, Carpellary lateral; V, Carpellary ventral).

lobes (Figs. 8–11). Within each lobe the sepal bundle gives out lateral branches on each side which show further branching (Figs. 10–12).

Close above the departure of the sepal traces five petal traces and five stamen traces depart from the central stele successively (Figs. 4, 5). The petal traces lie on alternate radii and the stamen traces on the same radii as those of the sepals. All these ten traces move out and form a ring within the cortex of the receptacle (Fig. 5). Simultaneously with the separation of the calyx tube, the corolla tube containing petal and stamen bundles is also separated from the receptacle (Figs. 6, 7). The petal bundles move up and each branches into a petal median and two petal laterals below the level of separation of the filaments (Fig. 11). The stamen traces pass out into the filaments and extend upto the connective (Figs. 11, 12). Just at this level the corolla tube breaks up into five lobes, each having a petal median and a petal lateral on either side (Fig. 11). The petal bundles divide further while extending up (Fig. 12).

Shortly after the departure of the stamen traces the receptacular stele breaks up into two halves, each half being trilobed. The median lobes move out and form carpellary dorsals. Each of the inwardly oriented lobes divides into a carpellary ventral that remains in the centre and a carpellary lateral which moves to a position on the side of a carpellary dorsal (Figs. 5–7).

The ovary is unilocular with two parietal placentae, two projecting ridges and four ovules (Fig. 7). The carpellary ventrals furnish ovular supply and are consumed. At this level the ovary appears to be deeply bilobed. Gradually toward higher levels, the two placentae as well as the two ridges meet in the centre thus making the ovary tetralocular (Fig. 8). At this level the ovary shows two dorsal wings at the back of the carpels (Fig. 9). The carpellary dorsals are situated within the wings. At a higher level the carpellary dorsals move into the central part of the ovary which later separates as gynobasic style (Fig. 10). The four ovary lobes also separate from one another. The carpellary lateral within each ovary lobe branches repeatedly and the branches anastomose variously. Only the carpellary dorsals continue upto the stigma (Figs. 11, 12). The stigma is broad at the base and tapering upward.

The floral anatomy of *Sericostoma pauciflorum* resembles in general with the vasculature described earlier by Joshi[1], Lawrence[2] and Sharma[3] for members of Boraginacaee. However, the vasculature of the sepals in this species is completely different from any other member of Boraginaceae described so far. The sepals are single traced and unlike in other members sepal bundles branch only within the free lobes of the calyx.

The vascular supply to the gynobasic style contains only carpellary dorsals as in other members. Two dorsal wings containing the carpellary dorsals are observed in this species. Lawrence[2] reported the presence of two dorsal wings in two species of *Alkanna*, four species of *Cynoglossum* and *Rindera ochroleuca* but in these species the dorsal bundles were reported to give out weak branches within the wings. No such branching has been observed in *Sericostoma pauciflorum*.

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---

1. Joshi, B. M., *Geobios*, 1975, 2, 10.
2. Lawrence. J. R., *Amer. J. Bot.*, 1937, 22, 433.
3. Sharma, R., *Agra Univ. J. Res.* (*Sci.*), 1954. 33, 349.

PROTECTION OF MATERIALS IN THE SEA

Symposium on deterioration of material in seawater and its prevention-marine corrosion and fouling will be held during February 25–26, 1977 at Bombay.

Details regarding the Symposium can be had from the Convener, Symposium Programme 1977, Naval Chemical and Metallurgical Laboratory, Tiger Gate, Bombay 400 023.

SHORT SCIENTIFIC NOTES

Occurrence of *Alepas* Rang (1829) in Indian Waters (Cirripedia, Lepadidae).

Alepas navigator, the pedunculate cirripede has been reported and described by Pilsbry[1], from Philippines on a Discomedusa, during the Philippine cruise of the 'ALBATROSS' 1907–1910.

During our investigation on the Cirripede fauna off the coast of Porto Novo (Lat. 11° 30′ N ; Long. 79° 49′ E), South India, a single specimen of *Alepas navigator* Pilsbry was found washed ashore during January, 1972. The present material appears to be the second find after a lapse of more than six decades.

The present material has the following dimensions. The length of the capitulum, 36 mm ; greatest width of the capitulum, 23 ; length of the peduncle, 16 ; diameter of the peduncle in the middle, 7.

We thank Dr. R. Natarajan, Director, for encouragement and Dr. Victor A. Zullo, University of North Carolina at Wilmington, U.S.A., and Dr. H. G. Stubbings, Admiralty Materials Laboratory, U.K., for help in identification. One of us (A. S. F.) thanks the U.G.C. for financial help.

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Annamalai University, K. Ramamoorthi.
Porto Novo 608 502,
Tamil Nadu, South India,
February 9, 1976.

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1. Pilsbry, H. A., *Proc. U.S. nation. Mus.*, 1912, 42, 291.

Coumarins of *Artemisia parviflora* Roxb.

Artemisia spp. have been reported to contain a number of coumarins and flavones[1]. The present study has been done using *Artemisia parviflora* Roxb. collected from the high altitudes of Western Himalayas in Laddak and Lahaul. Whereas, *Artemisia maritima*[2] is rich in santonin and is found in the valley (around Baramulla), *A. parviflora* has no santonin like *Artemisia scoparia*[3] found around Jammu Tawi, but contains three closely related coumarins, aesculetin, scopoletin and dimethyl ether of aesculetin.

Aerial parts (700 g) after repeated extraction with petroleum ether (60–80°) was exhaustively extracted with rectified spirit. The alcoholic concentrate (10·5 g) was chromatographed over neutral alumina (320 g) and eluted with solvents (100 ml) of increasing polarity (1) benzene (2) benzene + chloroform mixtures (*a*) 3 : 1, (*b*) 1 : 1, (*c*) 2 : 3 and (3) ethyl acetate yielding three substances A (1·55 g), B (0·90 g) and C (0·46 g). Substance 'A' on recrystallisation from ethyl acetate melted at 144–145°, molecular weight 206 (mass spectra), λ_{max}^{MeOH} 230, 250, 295, 345 (log $\epsilon = 4 \cdot 28, 3 \cdot 78, 3 \cdot 75, 4 \cdot 08$) $\nu_{max}^{(KBr)}$ 1732, 1635, 1575, 1480; and 870 cm^{-1}. The compound was found identical with 6, 7-dimethyl aesculetin. $R_f = 0 \cdot 90$ tlc, silica gel, solvent: BEA (benzene : ether, acetic acid, 87 : 12 : 1).

Substance 'B' (recrystallised from acetone) m.p. 200–202° λ_{max}^{MeOH} 229, 261, 295, 245 nm (log $\epsilon = 4 \cdot 25, 3 \cdot 8, 3 \cdot 83, 4 \cdot 2$) ν_{max} 3505, 1720, 1610, 1575, 1462, and 870 cm^{-1}, $R_f = 0 \cdot 77$ (tlc., *silica* gel., solvent : BEA). The substance was identified as scopoletin. Complete methylation with dimethyl sulphate in anhydrous acetone gave substance 'A'.

Substance 'C', after recrystallisation melts at 266–68° C, gives green coloration with 1% ferric chloride in ethanol, λ_{max}^{MeOH} 230, 355, $R_f = 0 \cdot 44$ (tlc, solvent BEA). Found identical with aesculetin. Complete methylation with dimethyl sulphate in anhyd. acetone gave 6, 7-dimethyl aesculetin.

Regional Research M. P. Jain.
Laboratory, R. S. Thakur.
Jammu–Tawi, P. R. Rao.
August 16, 1974.

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1. Chandrashekhar, V., Krishnamurti, M. and Seshadri, T. R., *Curr. Sci.*, 1965, p. 609.
2. Saha, S. K., *J. Ind. Chem. Soc.*, 1957, 20, 148.
3. Singh, G. B., Nayar, G. V. and Aggarwal, K. P., *Chem. and Ind.*, 1954, p. 1294.

Single Corpuscle of Stannius in the Teleost *Lepidocephalichthys guntea*

The corpuscles of Stannius are putative endocrine glands found in the mesonephros of holostean and teleostean fishes. The secretion (s) of corpuscles lack positive identification[1]. In a study of the renal anatomy of about 100 specimens of *Lepidocephalichthys guntea* two specimens with a single corpuscle of Stannius were encountered. Normally the corpuscles are absent in this species.

The kidney in *Lepidocephalichthys* is a fused organ and the single oval corpuscle is embedded in the posterior region. On the peritoneal side encapsulated by fibrous connective tissue, which also penetrates the corpuscle to divide it into distinct lobes. The rich vascular supply follows the collagenous septa. Each lobe contains loosely packed round cells free from connective tissue. The cells have prominent boundaries and are mononucleated. The slightly basophilic cytoplasm is scanty. A few large cells packed with granules stained with hematoxylin occur adjacent to the blood sinusoid.

This single corpuscle of Stannius in *Lepidocephalichthys guntea* is interesting since the minimum number so far recorded is two and the other members of this species lack any corpuscle. The histology of this single corpuscle is of type III[3].

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December 8, 1975.

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1. Chester Jones, I. *et al.*, *J. Endocrinol.*, 1965, 33, 319.
2. Krishnamurthy, V. C. and Bern, H. A., *Acta Zool.*, 1973, 54, 9.
3. — and —, *Gen. Comp. Endocrinol*, 1969, 13 (3), 313.

Search for Sources of Resistance to Powdery Mildew in Black-Gram

Powdery mildew (*Erysiphe polygoni* DC) of black-gram (*Phaseolus mungo* L.) occurs almost every year in varying intensity depending upon climatic conditions at the time of pod-formation and maturity of the crop in Uttar Pradesh. However, no information appears to be available, so far, on the varietal resistance to this disease. The present study was, therefore, taken up at Kanpur and the results of testing of 306 varieties and strains of black-gram in the adult stage under epiphytotic conditions for three years (1972, 1973 and 1974) are reported in this communication.

In September, 1972, the powdery mildew appeared in an epidemic form and some of the genotypes among the cultures of black-gram, sown at the University Farm, were found to show varying degrees of resistance. These varieties were subsequently tested for two years under conditions of artificial epiphytotics both in the field as well as in glass-house by the technique of Mains and Deitz[1] and observations on disease reactions were also recorded according to his scale with some modifications. The varieties were placed in various categories of resistance and susceptibility on the basis of type of reactions as follows:

Highly resistant (Macroscopically no mycelium is evident)	Nil
Resistant (Trace to slight development of fungus evident macroscopically)	6203
Moderately resistant (Light development of fungus occurs)	65-1, 5902-1 and 5905-1-1
Moderately susceptible (A moderate development of fungus occurs accompanied by moderate sporulation)	A-1, H 10, T 27, 5902, 5905-1, 6311, 6315 and 6403
Very susceptible (Abundant fungal growth is evident accompanied by abundant sporulation	294 varieties and strains including B 76, G1, L1 L35, L64, T9, T65, T77, UL1, UL2, UL131 and UPU2.

The results reveal that out of the 306 genotypes tested, only one proved resistant, three moderately resistant and the rest were found to be susceptible. The germplasm lines found resistant and moderately resistant may be utilized for breeding powdery mildew resistant varieties of black-gram.

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D. V. Singh.
R. R. Singh.
A. N. Mishra

May 11, 1975.

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1. Mains, E. B. and Deitz, S. M., *Phytopathology*, 1930, **20**, 229.

Asteromella: A New Generic Record from India

Asteromella morgan-jonii spec. nov.

Pycnidiis maculae foliicolae, gregaria, atro-brunnea, pusilus, globosa, vel subglobosa, subepidermalis, innata, postea erumpentia, ostiolatis, 70–130 μ diam, conidiophora hyalina, laevis, enteroblastic; conidiis hyalina, semel-cellularis, cylindricus vel bacillaris, 2–2·8 × 1·2–1·5 μ.

Ad foliis viventibus *Citrus maxima* (Burm.) Merril. (*C. decumana* L.), (Rutaceae), Pachpedi, Jabalpur, India, mense juli 1973, leg. N. D. Sharma.

Typus positus in Herb., I.M.I., Kew, sub numero, 186102, also in H.P.P., J.N.U. sub numero, 22.

The type specimen of the genus was sent to Commonwealth Mycological Institute, Kew, but it could not be placed under any of the existing species of *Asteromella*. So far no *Asteromella* has been reported on *Citrus*[3]. It is therefore described here as a new species. The species is described after Dr. G. Morgan Jones for his outstanding contribution in the group Coelomycetes (Fungi Imperfecti), *Asteromella morgan-jonii* spec. nov. Tilak[7] in his compilation wrongly indexed *Asterostomella strophanti*[1] P. Henn. as *Asteromella strophanti*. In fact *Asterostomella* is a member of the order Peltrasterales[2] with "picnostromas com himenio invertido" of the family Asterinothyriaceae with, 'micelio livre hifopodiado" with feoamerosporos, picnostromas radiados, dehiscencia estelar; sem hipostroma; whereas *Asteromella*[9] Pass. and Thum. is a spermatial state of *Mycosphaerella*. The genus is a new record from India[4-8].

Pycnidia in leaf spots, gregarious, dark brown, samll globose to sub-globose, subepidermal, innate, later erumpent, ostiolate, 70–130 μ in diam; conidiogenous cells hyaline, smooth, enteroblastic; conidia hyaline, 1-celled, cylindrical to bacillar, simple, 2–2·8 × 1·2–1·5 μ.

On living leaves of *Citrus maxima* (Burm.) Merril. (*C. decumana* L.) (Rutaceae), Pachpedi, Jabalpur, India, July 1973, leg. N. D. Sharma.

The author expresses his grateful thanks to Dr, G. P. Agarwal for encouragement. Thanks are also due to Mr. A. Johnston, Director and Dr. E. Punithalingam of the Commonwealth Mycological Institute, Kew, for help in the identification of the species.

Department of Plant Pathology, N. D. SHARMA.
J.N. Agricultural University,
Jabalpur–4, M.P.
March 10, 1976.

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1. Agarwal, G. P. and Sahni, V. P., *Mycopath. Mycol. Appl.*, 1964, **22**, 245.
2. Batista, A. C. and Ciferri, R., *Ibid.*, 1959, **11**, 102.
3. Knorr, L. C., *Pans*, 1973, **19** (3), 441.
4. Mukerji, K. G. and Juneja, R. C., *The Fungi of India*, (1962–72). Emkay Publications, New Delhi, 1974, p. 223.
5. Rangaswami, G., Seshadri, V. S. and Lucy Channamma, K. A., *Fungi of South India*, Univ. Agric. Sci., Bangalore, 1970, p. 187.
6. Tandon, R. N. and Chandra, S., *Supplement to the List of Indian Fungi*, Univ. of Allahabad Studies, 1964, p. 246.
7. Tilak, S. T. and Rao, R., *Second Supplement to the Fungi of India*, Marathwada University, Aurangabad, 1970, p. 314.
8. Vasudeva, R. S., *Fungi of India, Supplement–I.* (Sci. Res. Monogr.), I.C.A.R., New Delhi, 1962, p. 206.
9. Von Arx, J. A., *The Genera of Fungi Sporulating in Pure Culture*, Verlag, von J. Cramer, 3301 Lehre, 1970, p. 288.

New Records of Spiders as Predators of Maize Borer and Maize Jassid

The maize borer, *Chilo partellus* (Swinhoe) and maize jassid, *Zyginidia manaliensis* (Singh) are important pests of maize and jowar in India. While studying the field behaviour of maize borer during 1975, the maize borer and maize jassid were found to be preyed upon by eight species of spiders. The spiders, found preying on the respective insects, were collected from the field and their predatory habit was confirmed by feeding experiments in the laboratory.

In the case of maize jassid two species of spiders, *viz.*, *Oxyopes* sp. (Oxyopidae) and *Pardosa leucopalpis* Gravely (Lycosidae) were identified. These spider species preyed upon both the nymphs and adults of the maize jassid. In the case of maize borer, six species of spiders belonging to four different families were identified. These spiders entered the funnel of the plant in the field and fed on the younger instar (first and second) larvae of the maize borer. These were *Thomisus cherapunjeus* Tikader (Thomisidae); *Marpissa tigrina* Tikader and *Phidippus punjabensis* Tikader (Salticidae); *Araneus sinhagadensis* Tikader and *Araneus* sp (Argiopidae) and *Oxyopes pandae* Tikader (Oxyopidae).

Spiders as predators of maize borer larvae have earlier been reported by Singh *et al.*[2]. They reported 13 species of spiders belonging to seven families including *Araneus* sp., *Oxyopes pandae*, *Marpissa* sp. and *Phidippus* sp. Jandu[1] reported *Marpissa tigrina* Tikader to feed on citrus psylla, *Diaphorina citri* Kuwayama and to constitute 32% of the total spider population on citrus and *Oxyopes pandae* Tikader to feed on citrus whitefly, *Dialeurodes citri* Ashmd at Ludhiana. The present report of 6 species of spiders *viz.*, *Oxyopes* sp. and *Pardosa leucopalpis* Gravely on maize jassid and *Thomisus cherapunjeus* Tikader, *Marpissa tigrina* Tikader, *Phidippus punjabensis* Tikader and *Araneus sinhagadensis* Tikader on maize borer are the first records from India.

The authors are grateful to Dr. B. K. Tikader, Deputy Director, Western Regional Station, Zoological Survey of India, Poona-5, for the identification of spider species.

Department of Entomology, GURDIP SINGH.
Punjab Agricultural University, G. S. SANDHU.
Ludhiana. *March* 15, 1976.

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1. Jandu, M. K., *Thesis*, Punjab Agricultural University, Ludhiana (Unpublished), 1972.
2. Singh, B., Battu, G. S. and Atwal, A. S., *Indian J. Ent.*, 1974, **36** (2), (in press).

Sunnhemp Rosette: A New Virus Disease of Sunnhemp

Crotalaria juncea L. (Sunnhemp) is used for green manuring and for fibre production. In 1973, a severe mosaic disease, associated with filiformy and rosetting symptoms, was observed on the sunnhemp plants near Lucknow. The size and number of flowers per plant as well as seed formation was reduced. The seeds were smaller and deformed. The disease incidence was 40–80%.

The disease was mechanically transmitted to a few host plants only. The virus induced hypersensitive reaction in a few hosts but systemic symptoms as described earlier were produced on sunnhemp (*Crotalaria juncea* L.) only.

(*a*) *Cyamopsis tetragonoloba* L.

Local lesions appeared both on primary cotyledonary as well as on secondary trifoliate leaves after

2–3 days of virus inoculation in the form of small and black necrotic spots and were easily countable.

(*b*) *Chenopodium amaranticolor* Coste and Reyn.

Chlorotic local lesions appeared after 4–6 days of virus inoculation on inoculated leaves, these later on turned necrotic.

(*c*) *Vigna sinensis* Savi.

Chlorotic spots appeared 4–6 days after virus inoculation on primary cotyledonary and secondary trifoliate, inoculated leaves.

Besides these hosts, 84 representatives, *viz.*, from families Amaranthaceae, Apocynaceae, Chenopodiaceae, Cucurbitaceae, Cruicferae, Compositae, Cannabinaceae, Leguminosae, Polemoniaceae and Solanaceae, were inoculated, but neither they produced any type of disease symptoms nor the virus could be recovered on back inoculation to test plants.

Seed transmission of virus in sunnhemp plants, infected as seedlings, ranged from 5–20%. Virus was also found to be seed borne at a rate of 5 to 10% when plants were infected before flowering.

The insect vectors, if any, could not be found. *Myzus persicae* Sulz., *Aphis gossypii* Glover, *Aphis craccivora* Koch. and white flies, *Bemisia tabaci* Gen, did not transmit the virus.

The virus in undiluted buffered sap was inactivated at 93° C and at a dilution of 5×10^{-5}. The longevity *in vitro* was 54–56 days at 20–28° C.

A comparison between the host range, symptomatology and physical properties of the present virus and that of other viruses producing mosaic symptoms on Sunnhemp[1–10] revealed that the causal virus did not closely resemble to any of the above viruses. The causal virus of Sunnhemp mosaic[2–3] has lower dilution end point and thermal inactivation point and shows systemic reaction in various hosts. The virus occurring in Lucknow differs from Delhi strain in producing more severe symptoms on sunnhemp and in showing hypersensitive reaction in a few hosts.

Apparently, this is an undescribed disease of sunnhemp and appears to be caused by a severe strain of Sunnhemp mosaic virus.

Department of Botany,
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India, *March* 1, 1976.
H. N. VERMA.
L. P. AWASTHI.

1. Jenson, D. D., *Phytopathology*, 1950, **40**, 512.
2. Raychaudhuri, S. P., *Curr. Sci.*, 1947, **16**, 26.
3. —, *Proc. 35th Ind. Sci. Cong. B*, 1948, p. 124.
4. Nagaich, B. B., and Vashishtha, K. S., *Indian Jour. Microbiol.*, 1963, **3**, 113.
5. Bose, R. D. and Misra, S. D., *Ind. Jour. Agric. Sci.*, 1938, **8**, 417.
6. Klesser, P. J., *Bothalia*, 1961, **7**, 497.
7. Smith, K. M., *A Text Book of Plant Virus Diseases* (1972), 3rd edn., Longman Group Ltd., London.
8. Capoor, S. P., *Curr. Sci.*, 1950, **19**, 22.
9. Raychaudhuri, S. P., Nariani, T. K. and Das, C. R., *Indian Phytopath.*, 1962, **15**, 79.
10. — and Pathanian, P. S., *Curr. Sci.*, 1950, **19**, 213.

REVIEWS AND NOTICES OF BOOKS

Energy (Vol. II). *Non-nuclear Technologies.* By S. S. Penner and L. Icerman. (Addison-Wesley, Pub. Co., Inc., Mass. 01867), 1975. Price : $ 19.50 (Hardbinding), $ 13.50. Paperbinding.

The book under review is the second of a three-volume set on "Energy" and deals with non-nuclear Technologies. The topics covered are restricted to those aspects of non-nuclear energy technologies with new developments, especially in the U.S.A., which promise to bring important modifications to their energy supply base. Well-developed technologies are not reviewed.

The book is divided into twelve chapters—Chapters 9 to 20, the first eight chapters forming the contents of Volume 1.

Chapter 9 deals with the technological and economic aspects of oil recovery from tar sands and oil shale, Chapter 10 considers problems associated with the availability, production and transportation of coal, coal gasification, coal liquifaction and *in situ* recovery of coal. Chapter 11 is devoted to the use of hydrogen as an alternative primary energy source. Topics considered include production of hydrogen from water, hydrogen transmission and distribution networks, storage of hydrogen and safety problems connected with hydrogen use. Methanol as an alternative energy source is also briefly considered. Chapter 12 is concerned with energy storage systems such as mechanical energy storage, electrical energy storage, pumped-hydraulic storage, compressed air storage, storage of petroleum, natural gas and synthetic fuels. In Chapter 13 Techniques for Direct Energy conversion are descussed including MHD, fuel cells, photovoltaic power conversion, Thermoelectric power generation and thermionic power conversion. Chapter 14 deals with solar-energy utilisation while Chapters 15 and 16 relate to winomills and tidal energy. Chapter 17 discusses hydroelectric power generation with special reference to United States while Chapters 18 and 19 deal with Hydro-thermal Energy sources and Geothermal energy from Dry wells. The last chapter is devoted to Electrical Power Production, Transmission, and Distribution including application of cryoresistive cables and superconducting cables.

The authors are associated with the Energy Centre, University of California, San Diego and the book is a product of their lecture notes delivered to the UCSD graduate and undergraduate students. The reviewer has no hesitation in recommending this book as a text book for graduate students on "Energy" and the information presented therein will also be of great use to those involved in tackling the problems of energy with a view to reduce dependence from foreign energy supplies.

H. N. RAMACHANDRA RAO.

Development Biology—Pattern Formation Gene Regulation. Edited by Daniel McMahon and C. Fred Fox. (W. A. Benjamin, Inc., Menlo Park, Reading, Massachusetts), 1975, Pp. 604. Price $ 19.95.

Developmental Biology, Volume two, published by W. A. Benjamin, Inc., 1975, is compiled with the papers presented at the Conference on Developmental Biology, held in Squaw Valley, California, in March 1975. The articles cover basically two interdependent areas, namely, Cellular adhesion and pattern formation and Regulation of Translation and Transcription. As one of the investigators puts it, the main theme of the symposium appears to be how development can be explained in terms of genes controlling cell behaviour, and how cell behaviour can in turn determine supra-cellular organisation and morphogenesis. A wide variety of experimental systems ranging from slime moulds to higher animals have been used. *Dictyostelium discoideum* has been a favourite choice of the developmental biologists, where the development and differentiation processes have been studied with respect to the involvement of plasma membrane, cyclic AMP, changes in protein and transfer RNA and regulation of gene expression. *Drosophila* is another system where the availability of morphogenetic mutants has helped studies on differentiation. Efforts have been made to identify the nature of receptors involved in cell to cell recognition and interaction. The involvement of cellular position and its determination have been examined with relevance to the process of differentiation. In higher systems, penetrating studies have been carried out in understanding the molecular mechanism of action of hormones in eliciting the synthesis of specific proteins. Changes in nuclear RNA, poly adenylic acid containing RNA, chromatin and histones have been discussed with respect to gene activation and RNA turnover. Some of the other systems investigated are differentiation of the myogenic cell line, neuron differentiation, etc.

The volume contains 44 original articles covering the broad areas mentioned above. In addition, two articles are devoted to a study of abnormal human development with specific reference to purine metabolizing enzymes and cystinosis. The articles detail some of the latest research in the field of developmental and molecular biology and should be of immense value to advanced students and research workers in the field.

G. PADMANABAN.

Enzyme Reactions and Enzyme Systems. By Charles Walters. (Marcel Dekker, Inc., New York and Basel), 1976. Pp. xvi + 244. Price $ 19.50.

One is misled by the title. It is disappointing for a conventional enzymologist that the contents are on kinetics and mathematical modelling. In the author's own words the "purpose of this book is to describe how the catalytic properties of enzymes and multienzyme systems can be translated into mathematical models, and how these models, when compared with experimental observations, can be used to deduce information about chemical mechanisms of enzyme catalysis". The book seems to serve less than that. There are only 40 references given at the end, of which 16 belong to the author. It is a reflection of how narrow is the treatment for such a broad title. Such specialized monographs are best fitted for series reviews. The book covers the following aspects : The general theory of enzyme kinetics, the quasi-steady-state assumption, Quasi-steady-state enzyme models, Simulation of enzyme models, Analysis of enzyme kinetic data, Theory of control, Chemical kinetics of multicomponent systems without feedback, and Chemical kinetics of multicomponent systems with feedback control. An enzymologist craving for work on kinetics and computor or a systems scientists plung-
kinetics and computer or a systems scientist plung-

T. RAMASARMA.

Annual Review of Physiology (Vol. 38). Editor: Ernst Knobil. Associate Editors: Ralph R. Sonnenschein and I. S. Edelman. (Annual Reviews, Inc., Palo Alto, California, U.S.A.), 1976. Pp. viii + 649. Price: U.S.A. $ 17.00. Elsewhere: $ 17·50.

The ever expanding, shifting and dissolving boundaries of physiology render the selection of the topics to be reviewed increasingly difficult. In this Annual Review, an attempt has been made to focus attention on some topics of current interest.

Renal physiology highlights four topics:

(i) mechanism of glomerular filtration and determinants of glomerular filtration rate, (ii) advances in the electrophysiology of the nephron, (iii) Renal tubular chloride transport and the mode of action of some diuretics and (iv) advances in renal tubular biochemistry.

The topics covered in "Hypothalamic hormones" include extrahypothalamic distribution, biosynthesis and degradation and neurotransmitter control of the hypophysiotrophic hormones and physiological aspects of a newly discovered hypophysiotrophic hormone—'The growth hormone release-inhibiting hormone, Somatostatin'. The review on higher functions of the nervous system concentrates on cognitive and elaborative mechanisms, electrophysiological correlates of attention, perceptual mechanisms, the motor elaboration systems, higher levels of integration, learning and plasticity. Increasing evidence for the existence of neurotrophic (non-impulse) mechanisms, especially in nerve-muscle cell relations, has been presented. 'Gating in Sodium Channels of nerve' summarizes some of the major new observations on gating mechanisms involved in the excitability of nerves. Neurotransmitter receptors in the brain have been successfully identified for most of the known neurotransmitters, utilising typical biochemical protocols. The role of the pyramidal tract neurons and other pre-central cells in modifying the pyramidal command movement performance by a feedback mechanism is reviewed.

Models of steroid hormone action, currently most in vogue, are critically examined, with special emphasis on the mechanism of action of estrogens. Recent biochemical work on the developmental processes in the preimplantation mammalian embryo related to the nature of control mechanisms, the role of the transport mechanisms and the effects of metabolic pools and compartmentalization are presented. 'Mass transport across cell membranes' reviews the effects of antidiuretic hormone on nonelectrolyte permeation, Na^+ transport and water flows in hormone sensitive epithelia. Regulation of insulin and glucagon secretion by substrates, neural and hormonal influences modulating the islets functions and the molecular interrelationship of hormone secretion are discussed.

Overall cardiovascular regulation reviews some important cardiovascular homeostatic reflex mechanisms with emphasis on the efferent pathways, as also the basic concepts on the superimposed remote control systems, both neuronal and hormonal. Failure of the body suffering from shock to restore the homeostatic equilibrium has been attributed to the inadequacy of the central nervous servocontrol system in hypovolemic and other types of shock. The review on platelets summarizes some of the recently developed concepts and data relating to the reaction of the platelet with its microenvironment.

In the study on excitation-contraction coupling, attention has been focussed on the question of how depolarization of the T system induces the release of Ca^{2+} from the sarcoplasmic reticulum. Biochemical adaptations deal with endurance exercise in muscles and the physiological consequences of these adaptations.

Mechanisms of heat production in mammalian cells with emphasis on their control mechanisms at cellular level are considered under cellular thermogenesis.

M. SIRSI.

ANNOUNCEMENTS

Symposium on General and Comparative Endocrinology

A Symposium on General and Comparative Endocrinology, sponsored by the Universities of Delhi and Karnataka and other Institutions interested in Endocrinology and Diabetes, will be held in Delhi from October 25–30, 1976.

Further information can be obtained from the convener, Prof. B. I. Sunderaraj, Department of Zoology, University of Delhi, Delhi 110 007.

Diabetic Association of India, Bombay

Dr. R. V. Sathe Endowment Award of Rs. 500/– will be awarded annually (January 1977) to a Post-Graduate below the age of 35 years, who has contributed to the knowledge of Diabetes in India.

Further details can be had from: Dr. A. S. Godbole, Hon. Secretary, Diabetic Association of India, 127, Mahatma Gandhi Road, Bombay 400 023.

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Collett, H., *Flora Simlensis.* Rep. 1971 pp. xxxii + 652 (Many illus.). Price Rs. 85·00 or $ 16.00.

Don, D., *Prodromus Florae Nepalensis.* Rep. 1976, 256 pp. Price: Rs. 75·00 or $ 19.00.

Duthie, J. F., *Flora of the Upper Gangetic Plain and of the Adjacent Siwalik and Sub-Himalayan Tracts.* Rep. 1973. pp. xxvii + 1050. Vols. 1–3. Pts. 1–2 (Bound in 2 vols.). Set Price: Rs. 60·00 or $ 15.00.

Dymock, W., *Pharmacographia Indica.* Rep. 1976. 3 Volumes 1914 pp. Set Price: Rs. 700·00 or $ 175.00.

Fyson, P. F., *Flora of Nilgiri and Pulney Hill Tops.* Rep. 1974–75, 3 Vols. pp. xlvi + 1342. Set Price: Rs. 575·00 or $ 100.00.

Gamble, J. S., *A Manual of Indian Timbers.* Rep. 1972, pp. xxvi + 868 (21 Illustrations), Price: Rs. 95·00 or $ 18.00.

Griffith, W., *Icones Plantrum Asiaticarum.* Rep. 1976, 8 Parts. 4 Parts of Text, 1900 pp. 4 Parts of Plates, Ab. 1161 Plates. Set Price of 8 Parts: Rs. 2500·00 or $ 675.00.

Hooker, J. D. and Thomson, T., *Flora Indica.* i-xv, 1–280, 1–285 pp. Rep. 1976. Price Rs. 175·00 or $ 35.00.

Hooker, J. D., *Flora of British India.* Rep. 1973. 7 Vols. pp. xl + 5568. Set Price: Rs. 750·00 or $ 130.00.

Hope, C. W., *Ferns of North Western India.* Rep. 1976. Price Rs. 85·00 or $ 19.00.

Kachroo, P. (Ed.), *Recent Advances in Botany,* (Prof. P. N. Mehra commemoration volume). Pub. 1976 Ab. 450 pp. Price: Rs. 95·00 or $ 19.00.

Kirtikar, K. R. and Basu, B. D., *Indian Medicinal. Plants.* Rep. 1975. 8 Volumes. 4 Vols. of Text, 2793 pp. 4 Vols. of Plates. 1033 Plates. Set of 8 Vols. Rs. 3000·00, £ 300.00 or $ 600.00.

Kurz, S., *Forest Flora of British Burma.* Rep. 1974 2 vols. pp. xxx + 1162. Price: Rs. 300·00 or $ 60.00.

Nairne, A. K., *Flowering Plants of Western India.* Rep. 1976. 401 pp. Price: Rs. 75·00 or $ 18.00.

Parker, R. N., *A Forest Flora for the Punjab with Hazara and Delhi.* Rep. 1973, pp. xxxvi + 578. Price: Rs. 150·00 or $ 30.00.

Parkinson, C. E., *A Forest Flora of the Andaman Islands.* Rep. 1972 pp. xxxvi + 326 with 6 plates. Price: Rs. 45·00 or $ 9.00.

Raizada, M. B., *Supplemnt to the Flora of the Upper Gangetic Plain and of the Adjacent Siwalik and sub-Himalayan Tracts.* By J. F. Duthie. Pub. 1976, Ab. 350 pp. (In Press).

Stewart, R. R., *Flora of Ladak, Western Tibet.* Rep. 1973 pp. 80, Price: Rs. 20·00 or $ 4.00.

Strachey, R., *Catalogue of the Plants of Kumaon and of the Adjacent portions of Garhwal and Tibet.* Rev. by J. F. Duthie, Rep. 1974, pp. vii + 269. Price Rs. 50·00 or $ 10.00.

Talbot, W. A., *Forest Flora of the Bombay Presidency and Sind.* Reprinted with CORRECTION AND NOMENCLATURAL CHANGES. 21 pp. by M. B. Raizada. Rep. 1975. 2 Vols. 1153 pp. (Many text illus.) Set Price: Rs. 300·00 or $ 70.00.

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Trimen, H., *A Handbook to the Flora of Ceylon.* Rep. 1974, Vols. 1–6 including 1 vol. of plates. Set Price: Rs. 1150·00, or $ 225.00.

Watt, G., *A Dictionary of the Economic Products of India.* Rep. 1972. 10 pts. in 6 Vols. *plus* Index Vol. Set Price: Rs. 1150·00 or $ 259.00.

Wight, R. and G. A. W. Arnott, *Prodromus Floreae Peninsulae Indiae Orientalis.* Rep. 1976. Price: Rs. 150–00 or $ 30–00.

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STEREOCHEMISTRY OF THE PYRROLIDINE RINGS IN THE COLLAGEN STRUCTURE

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ABSTRACT

In the collagen triple-helical structure, large side groups occurring at location 3 in the repeating triplet sequence (Gly–R_2–R_3)$_n$ are appreciably constrained if a proline residue occurs as R_2 in a neighbouring chain. The severity of the steric hindrance depends on the geometry of the prolyl ring. In this paper we propose two different puckerings for the proline ring, the first one being energetically favourable for most types of residue sequences commonly found in collagen, while the second is preferable when an amino acid residue with a large side group occurs at location 3 in a neighbouring chain. The puckering of the pyrrolidine ring of hydroxyproline, as proposed earlier, is quite favourable from energy, as well as stereochemical considerations.

THE primary structure of the fibrous protein collagen has the repeating tripeptide sequence (Gly–R_2–R_3), where R_2 and R_3 are, quite frequently, the imino acids proline and hydroxyproline respectively[1]. The secondary structure is a coiled-coil triple-helix with one direct inter-chain hydrogen bond per tripeptide, which links the amino group of a glycine residue in one chain and the carbonyl oxygen of the residue R_2 in a neighbouring chain[2]. A second set of hydrogen bonds can be formed, *via* a water molecule as an intermediary, in regions where the residue R_2 is not an imino acid[3]. In what follows, we shall refer to the residues R_2 and R_3 as being in ' locations ' 2 and 3, respectively.

We have recently examined the collagen structure in detail to see if there are any stereochemical restrictions on the occurrence of certain amino acids in the positions R_2 and R_3[4]. It was found that the presence of a proline residue as R_2 restricts the freedom of orientation of large side groups occurring as R_3 in a neighbouring chain. However, recent data (from theoretical energy calculations as well as from experiments) have shown that the pyrrolidine ring is not a rigidly fixed entity, but can take up a number of different ring puckerings[5–8]. We have therefore examined the possible geometries of the prolyl rings in the collagen structure, from stereochemical and energy considerations. It is found that the ring puckering for a proline residue at location 2, as given in Ref. 2, is slightly unfavourable if the hydrogen atoms of the methylene groups are explicitly taken into consideration, instead of regarding the methylene groups as single atoms of larger van der Waals radius, as was done earlier. In this paper we propose a slightly different geometry for the proline side chain, which does not have this bad feature. The most favourable ring puckering corresponds to one having type B conformation in the notation of Ref. 5. The C^δ atom is in the plane of the peptide unit and the ring has a C^γ-endo puckering. There are no bad contacts, involving the backbone atoms, less than the extreme limit of the contact criteria[9]. The ring puckering is also very favourable from energy considerations (as can be seen from Table II of Ref. 5). There is only one H ··· H contact of the order of 1·87 Å.

A projection of the collagen triple helix down the helical axis, with a prolyl residue at location 2, is shown in Fig. 1. The various bond angles and dihedral angles describing the geometry of the prolyl ring are given in Table I (first row) and the cylindrical polar coordinates of the atoms of the proline residue (in chain A) are given in Table II. The hydrogen atoms have been fixed in a tetrahedral orientation at the various carbon atoms, with the C—H bond length being fixed at 1·1 Å.

The proline ring with this conformation does not cause any steric hindrance to the occurrence of any other amino acid residue at location 3 in the same chain. But as can be seen from Fig. 1, the side group of a residue at location 3 in chain A is oriented towards the prolyl residue at location 2 in chain B. Hence large side groups occurring in this location in chain A are appreciably constrained in their freedom of orientation. However, all side groups, except those with branching at C^γ (as in leucine and phenylalanine residues) can occur at location 3, though with a limited range of allowed orientations. A side chain, branched at C^γ, can also be accommodated at location 3, in the presence of a prolyl residue at location 2 in a neighbourng chain, if the puckering of the proline ring is slightly altered.

The most favourable geometry of the prolyl ring, if large side groups occur at location 3 in a neighbouring chain, is shown in Fig. 1 by dotted lines. The C^δ atom has been moved about 14° out of the plane of the peptide unit and the C^γ atom is in an exo position. This corresponds to a type A conforma

TABLE I

Various parameters which describe the geometry of the pyrrolidine rings of proline and hydroxyproline residues in the collagen structure as described in this paper. The notation follows Ref-5+

Residue	τ^{α}	τ^{β}	τ^{γ}	τ^{δ}	τ^{N}	θ	χ^1	χ^2	χ^3	χ^4
(*a*) Pro	103	107	106	104	113	−9	22	−26	20	−7
(*b*) Pro*	105	107	106	105	111	+5	−18	25	−22	10
(*c*) Hyp†	106	106	105	104	111	−5	−14	26	−29	21

+ The τ's denote the internal bond angles of the pyrrolidine ring, while the dihedral angles correspond to: θ (C^{β}—C^{α}—N—C^{δ}), χ^1 (N—C^{α}—C^{β}—C^{γ}), χ^2 (C^{α}—C^{β}—C^{γ}—C^{δ}), χ^3 (C^{β}—C^{γ}—C^{δ}—N) and χ^4 (C^{γ}—C^{δ}—N—C^{α}).

* This geometry of the proline ring is preferable when large side groups occur in a neighbouring chain (see text).

† These values are for the Hyp ring as given in Ref. 2, and found to be satisfactory in this investigation,

tion of Ref. 5. The various parameters describing its geometry are given in Table I (second row) and the coordinates of the relevant atoms are given in Table II.

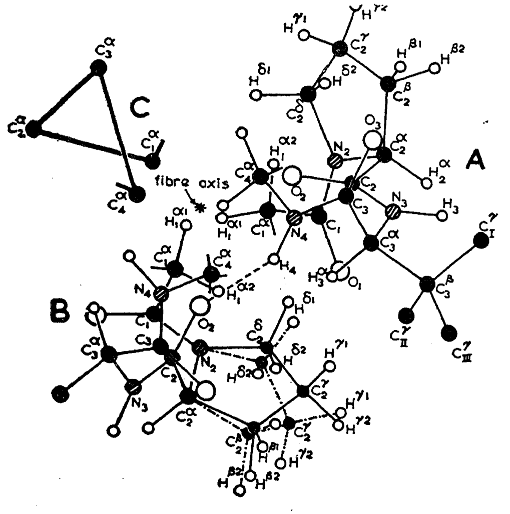

FIG. 1. A projection of the collagen triple-helix down the helical axis. A proline residue is shown at location 2, while C^{γ} atoms, in all the three possible staggered orientations, are shown attached to C_3^{β} in chain A. The puckering of the proline ring, which facilitates the occurrence of large side groups at location 3 in chain A, is shown by broken lines.

TABLE II

The atomic coordinates, in a cylindrical polar system for a proline residue at position 2 in chain A are given for the two proposed geometries of the proline ring (listed in Table I). The coordinates for a proline residue in chain B (shown in Fig. 1) can be obtained by a transformation using the helical parameters (*unit height* = 2·91 Å, *unit twist* = − 110)°

Geometry	Atom	r (Å)	ϕ (°)	z (Å)
Common atoms	N_2	2·96	21·90	1·62
	C_2^{α}	3·95	18·20	2·70
	C_2	3·17	11·50	3·96
As in row (*a*) of Table I	C_2^{β}	4·83	34·55	2·79
	C_2^{γ}	4·40	50·60	2·20
	C_2^{δ}	3·27	47·60	1·20
	H^{β_1}	5·10	36·70	3·85
	H^{β_2}	5·75	31·70	2·24
	H^{γ_1}	4·20	60·80	2·95
	H^{γ_2}	5·32	54·40	1·65
	H^{δ_1}	2·65	65·50	1·26
	H^{δ_2}	3·64	46·60	0·17
As in row (*b*) of Table I	C_2^{β}	4·85	34·28	2·80
	C_2^{γ}	4·86	43·97	1·57
	C_2^{δ}	3·45	44·59	1·00
	H^{β_1}	4·66	42·07	3·65
	H^{β_2}	5·88	29·90	2·96
	H^{γ_1}	5·26	55·52	1·82
	H^{γ_2}	5·57	38·76	0·85
	H^{δ_1}	3·12	62·27	1·29
	H^{δ_2}	3·51	42·77	−0·08

The energy of the triple-helical structure for this geometry of the prolyl ring is only about 2 kcal/mole per tripeptide higher than for the other geometry (but this will be made up by the packing energy of the large side chains). The values for the intra- and inter-chain energy (nonbonded + electrostatic) for the tripeptide sequence (Gly–Pro–Ala)$_n$ for the two different ring puckerings are given in Table III, along with the short contacts which are less than the extreme limits. The energy values for the conformations of the triple-helix, as given by other workers[10, 11], are also given in Table III. They are similar to the values for the Ramachandran one-bonded model. In these models also, the proline ring has the type B conformation and causes appreciable steric hindrance to the occurrence of large side groups at location 3 in the neighbouring chain.

TABLE III

The intra- and inter-chain energy values (nonbonded + electrostatic) in kcal/mole per tripeptide for the repeating tripeptide sequence (Gly–Pro–Ala)$_n$, with the triple-helical conformation proposed by various workers. The constants used to calculate the nonbonded and electrostatic contributions have been taken from Refs. 9 *and* 13 *respctively*

Structure	Energy (nonbonded + electrostatic) in kcal/mole			Short contacts	
	Intra-chain	Interchain	Total	Type	Distance (Å)
Ramachandran 1-bonded, as in Ref. 2, but with new proline geometry, (*a*) of Table I	−17·14	−13·58	−30·72	H_3^{α} (A) ··· $H^{\gamma 1}$ (B)	1·87*
Same as above, but with proline geometry, (*b*) of Table I	−16·18	−12·39	−28·57	$H_1^{\alpha 2}$ (A) ··· $H^{\delta 2}$ (A)	1·83
Yonath and Traub as in Ref. 11, but with C—H bond length equal to 1·1 Å	−17·10	−11·59	−28·69	H_3^{α} (A) ··· $H^{\delta 1}$ (B)	1·77*
Rich and Crick (II) as in Ref. 10, but with hydrogens also attached	−17·90	−13·36	−31·32	..	..

* Denotes short contacts involving the proline ring atoms in a neighbouring chain.

Thus, by allowing small variations in the puckering of the proline ring, it is possible to accommodate any amino acid at location 3 even in the presence of a prolyl residue at location 2 in a neighbouring chain. Hence any polytripeptide with a sequence of the type (Gly-Pro-R_3)$_n$ can take up a collagen-like triple-helical structure, even if residue R_3 has a bulky side chain, as is found to be the case from studies on synthetic polytripeptides[11, 12].

The geometry of the pyrrolidine ring of the hydroxyproline residue, as given in Ref. 2, is quite favourable energetically. It has a type A conformation with C^{γ} exo, the ring geometry being very similar to that found in crystal structures of 4-hydroxy-L-proline[14, 15] and in solution for poly (L-hydroxyproline)[16]. The bond angles and dihedral angles describing the ring geometry are given in Table I, row (*c*). There is only one bad contact involving a hydrogen of the δ-methylene group ($H_3^{\delta} \cdots H_2^{\alpha} = 1\cdot76$ Å). The presence of a hydroxyproline residue at location 3 slightly restricts the freedom of orientation of a side group R_2 in the same chain, if the latter occurs with its C^{γ} atom *trans* to the N atom, about the bond C^{α}—C^{β}. However these restrictions are not very severe. The hydroxyproline residue also does not cause any appreciable steric hindrance to the occurrence of any amino acid residues in the neighbouring chains. It may be mentioned that the hydroxyproline ring, with this puckering, is suitably oriented so as to form a hydrogen bond with the water molecule linking the two chains of the triple-helix, as well as an inter-protofibrillar hydrogen bond with a neighbouring triple-helix, thus giving additional stability to the collagen structure[17],

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1. Hulmes, D. J. S., Miller, A., Parry, D. A. D. Piez, K. A. and Woodhead-Galloway, J., *J. Mol. Biol.*, 1973, 79, **137**.
2. Ramachandran, G. N., In *Treatise on Collagen* (G. N. Ramachandran ed.), Academic Press New York. 1967, **1**, 130.
3. — and Chandrasekaran, R., *Biopolymers*, 1968, **6**, 1649.
4. Bansal, M., *Int. J. Pept. Prot. Res.*, 1976 (in press).
5. Ramachandran, G. N., Lakshminarayanan, A. V., Balasubramanian, R. and Tegoni, G., *Biochim. Biophys. Acta*, 1970, **221**, 165.
6. Venkatachalam, C. M., Price, B. J. and Krimm, S., *Macromolecules*, 1974, **7**, 212.
7. Balasubramanian, R., Lakshminarayanan, A. V., Sabesan, M. N., Tegoni, G., Venkatesan, K. and Ramachandran, G. N., *Int. J. Protein Res.*, 1971, **3**, 25–33.
8. Torchia, D. A. and Lyerla, J. R., Jr., *Biopolymers*, 1974, **13**, 97.
9. Ramachandran, G. N. and Sasisekharan V., In *Advances in Protein Chemistry* (Anfinsen, C. B. Jr., Anson, M. L., Edsall, J. T. and Richards, F. M., eds.), Academic Press, New York, 1968, **23**, 284.
10. Rich, A. and Crick, F. H. C., *J. Mol. Biol.*, 1961, **3**, 483.
11. Traub, W. and Piez, K. A., In *Advances in Protein Chemestry* (Anfinsen, C. B. Jr., Edsall, J. T. and Richards, F. M., eds.), Academic Press, New York, 1971, **25**, 243.
12. Scatturin, A., Tamburro, A. M., Del Pra, A. and Bordingnon, E., *Int. J. Pept. Prot. Res.*, 1975, **7**, 425.
13. Poland, D. and Scheraga, H. A., *Biochemistry*, 1967, **6**, 3791.
14. Donohue, J. and Trueblood, K. N., *Acta Cryst.*, 1952, **5**, 419.
15. Koetzle, T. F., Lehmann, M. S. and Hamilton, W. C., *Ibid.*, 1973, **29 B**, 231.
16. Torchia, D. A., *Macromolecules*, 1972, **5**, 566.
17. Bansal, M., Ramaksrihnan, C. and Ramachandran, G. N., *Proc. Indian Acad. Sci.*, 1975, **82 A**, 152.

SOLVENT EFFECT ON THE ULTRAVIOLET ABSORPTION SPECTRA OF ORTHO- AND PARA-NITROANISOLES

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A GENERAL programme has been undertaken in this laboratory to investigate the absorption and emission spectra in solution in solvents with a view to using the results to determine the dipole moments of molecules in their electronic excited states. As a first step in this programme, the authors have studied the absorption spectra of ortho- and para-nitroanisoles in different solvents and the results are reported and discussed.

Following Suppan[2] the magnitude and direction of the solvent shift of the absorption band between solvents *a* and *b* with nearly equal refractive index is given by the equation

$$-\triangle E_{a-b} = \frac{(\vec{\mu}_e - \vec{\mu}_g)\cdot}{a_g{}^3}\vec{\mu}_g \triangle f(D)_{a-b} \qquad (1)$$

where $\triangle E_{a-b}$ is the solvent shift in the absorption band between the solvents *a* and *b*, $\triangle f(D)_{a-b}$ is the difference between the $f(D)$ values corresponding to the solvents *a* and *b* where $f(D) = 2(D-1)/(2D+1)$ (D is the dielectric constant), μ_e is the dipole moment of the solute molecule in the excited state of the molecule. μ_g is the dipole moment of the solute molecule in the ground state, a_g, is the cavity radius of the molecule in the ground state and is assumed to be equal to the cavity radius of the molecule in the excited state, assuming that the molecular volume does not change appreciably on excitation.

The absorption spectra of pure samples of para-nitroanisole in cyclohexane, carbontetrachloride, decalin, *n*-heptane-, *n*-butyl acetate, *n*-amyl acetate and tetrahydrofuran and of ortho-nitroanisole in *n*-heptane, *n*-butyl acetate, *n*-amyl acetate, tetrahydrofuran, cyclohexane, benzene and toluene have been recorded on Beckmann DK2A spectrophotometer. The maximum of absorption band for each of the two solutes corresponding to the first excited state has been located in different solvents and thus the values of the $\triangle E$ have been obtained.

The static dielectric constants of the pure solvents have been measured at 1 MHz and at 25° ± 1° C with the help of a circuit comprising Franklin Oscillator-wavemeter combination.

The refractive indices for the sodium line of the solvents have been measured with an Abbe' type refractometer and the values are accurate up to the third decimal place. The dipole moments of the solutes have been determined in dilute solutions using the modified Guggenheim's method[6]. The cavity radius in each solute has been assumed to be approximately equal to the molecular radius of the solute molecule and has been estimated in each case by calculating the molecular volume using atomic increatments[7] instead of using the equivalent shell method[2].

The measured values of the dielectric constants D and refractive indices *n* of the solvents used in the present investigation, the estimated values of the functions,

$$\phi(D) = \frac{(D-1)}{(D+2)} \text{ and } f(D) = \frac{2(D-1)}{(2D+1)}$$

and the maximum of absorption band $\nu_{\max}$ (cm^{-1} corresponding to each solvent for the two solute molecues are collected in Table I. The experimentally determined ground state dipole moment values μ_g, the estimated values of the cavity radius a_g and the dipole moments μ_e obtained in this study from equation (1) for the two solute molecules are given at the foot of this table. The plots of solvents shift *versus* $f(D)$ and $\phi(D)$ functions for the two solute molecules are shown in (Figs. 1 and 2).

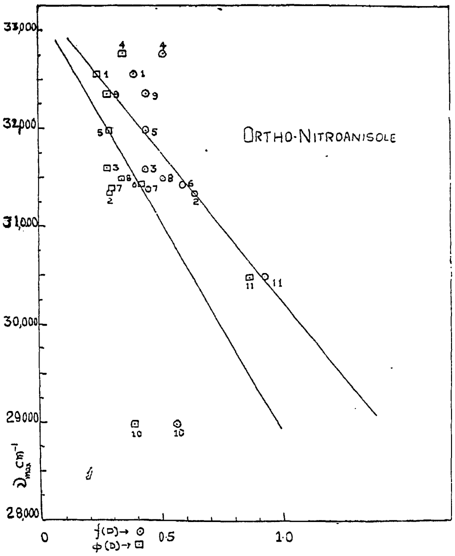

FIG. 1

Both the plots $\nu_{\max}$ *versus* $f(D)$ and $\nu_{\max}$ *versus* $\phi(D)$ for both the solute molecules lie on reasonably good straight lines and seem to show

TABLE I

Solvent shifts of ortho- and para-nitroanisoles (band S_1)

Solvents	D	f (D)	ϕ (D)	n	ν_{max} in cm^{-1}	
					ortho-nitro-anisole	para-nitro-anisole
1. *n*-heptane	1·96	0·390	0·242	1·401	32563	32458
2. *n*-butyl acetate	2·27	0·639	0·298	1·395	31338	31939
3. *n*-amyl acetate	2·17	0·438	0·281	1·400	31586	32458
4. Cyclohexane	2·58	0·513	0·345	1·425	32777	32510
5. Carbontetrachloride	2·22	0·449	0·289	1·459	31990	32353
6. Tetrahydrofuran	3·22	0·597	0·425	1·408	31420	31787
7. Benzene	2·28	0·461	0·299	1·499	31388	..
8. Toluene	2·56	0·510	0·342	1·495	31487	..
9. Decalin	2·17	0·438	0·281	1·476	32353	32300
10. Dioxane	2·95	0·565	0·394	1·409	28893	31289
11. Acetone	21·00	0·933	0·875	1·362	30479	30294

$\mu_g = 4\cdot83$ D; $a_g = 3\cdot043$ A; $\mu_g = 4\cdot95$ D; $a_g = 3\cdot043$ A
$\mu_e = 17\cdot9 \pm 4\cdot0$ D; $\mu_e = 23\cdot8 \pm 1\cdot0$ D.

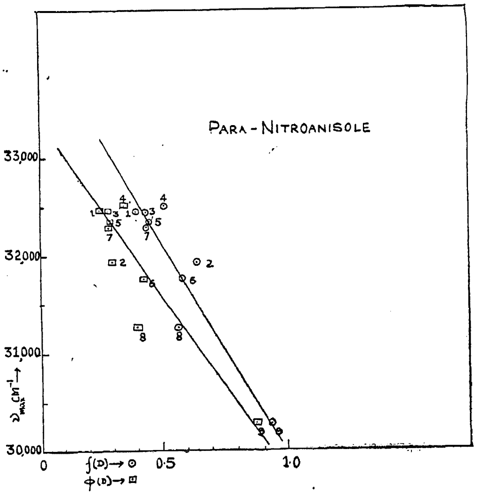

FIG. 2

less scatter than those molecules reported in the literature[1-4]. The change in either f (D) or ϕ (D) with ν_{max} of different solvents seems to be smaller and correspondingly, the plots are steeper. Therefore the change in the dipole moment upon electronic excitation is larger and therefore the dipole moment in the excited state is rather large for these molecules compared with those reported in the literature.

The ground state moment values for the two solute molecules increase from ortho-isomer to para-isomer. This may indicate that the positive end of methoxy dipoles in these isomers is towards the benzene ring, since it is known that the nitro group is symmetrical with its resultant moment acting in the direction of the group valence bond. However, the difference in the moments of these two isomers is not well outside the experimental errors. But the increase in the dipole moment from the ortho-isomer to the para-isomer in the excited state seems to support this point. The dipole moment of the para-isomer in the ground state as well as in the excited state is larger than that of the ortho-isomer which may mean that in the former molecule there is more transfer of charge into the ring than in the **latter.**

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1. Ledger, M. B. and Suppan, P., *Spectrochimica Acta,* 1967, 23 A, 641.
2. Suppan, P., *J. Chem. Soc.,* 1968, 10, 3125.
3. Favini, G., Gamba, A., Grasso, D. and Millefiori, S., *Trans. Far. Soc.,* 1971, 67, 3139.
4. Grasso. D.. Millefiori, S. and Fasone, S., *Spectrochimica Acta,* 1975, 31 A, 187.
5. Basu, S., *Adv. Quantum Chem.,* 1964, 1, 145.
6. Guggenheim, E. A., *Trans. Far. Soc.,* 1949, 45, 714 and 1951, 47, 573.
7. Edward, J. T., *Chem. and Ind.,* 1956, p. 774.

OXIDATION OF VALINE, LEUCINE AND PHENYL ALANINE BY CHLORAMINE-T

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ABSTRACT

Chloramine–T oxidises valine, leucine and phenyl alanine in various solvent media and the reaction is stoichiometric with a four electron change in buffer medium of pH 4–6. Back titration procedures for the estimation of these essential amino-acids in a buffer medium of pH 5 have been described. The products are the corresponding nitriles and *p*-toluene sulphonamide. The effect of foreign ions and other amino-acids on the oxidation has been investigated.

SEVERAL applications of chloramine–T (CAT) as an oxidant in the estimation of compounds of biochemical origin have been made recently[1,2]. In the present investigations, we have examined the behaviour of CAT as an oxidimetric reagent for the estimation of valine, leucine and phenyl alanine (PAN). It was found that these essential amino-acids are oxidized by CAT at room temperature in several solvent media. However, a stoichiometric oxidation with a 4–electron change is noticed at pH 5. Back titration procedures had to be adopted as the reaction was not fast enough for a direct titration with a visual or potentiometric end-points.

MATERIALS AND METHODS

DL-valine (BDH), leucine (Diamalt—West Germany) and DL-β-phenyl alanine (BDH), were recrystallized from aqueous medium and the purity was checked by the standard acetous perchloric acid method[3]. Chloramine-T (E'Merck) was purified by the method of Morris *et al*[4]. and the solution was standardised by the iodometric method. Reagent grade chemicals were used in preparing other solutions.

Preliminary experiments indicated that one m. mole of the amino-acid could consume 2 m. moles of CAT at pH 4–6 only. At other pH values, lower values were obtained, Even in presence of mineral acids (HCl, H_2SO_4 and $HClO_4$, 0·25–1·00 M), this ratio was considerably lower.

RECOMMENDED PROCEDURE

Solutions of the amino-acid (0·1–3·0 m. moles) in acetate buffer of pH 5·0 were pipetted to a measured excess (10–20%) of 0·1 N CAT. The reaction mixture was set aside for 30–40 minutes and the unconsumed CAT was estimated iodometrically. A blank titration was carried out with the same volume of CAT.

Some results of analyses are given in Table I. The results are accurate within 0·5%.

TABLE I

Estimation of valine, leucine and phenyl alanine by chloramine–T in pH 5 buffer medium
Amounts are expressed in mg.

Valine			Leucine			Phenyl alanine		
Taken	Found	% Error	Taken	Found	% Error	Taken	Found	% Error
2·00	2·01	+0·50	4·00	3·99	−0·25	4·00	4·01	+0·25
4 00	4 01	+0 25	6·00	6·01	−0·17	6·00	6·01	+0·17
6·00	5·99	−0·17	8·00	8·02	+0·25	8·00	8·02	+0·25
8·00	8·00	0·00	10·00	10·00	0·00	10·00	10·02	+0·20
10·00	10·01	+0·10	12·00	12·02	−0·17	20·00	19·98	−0·10
12·00	12·00	0·00	15·00	14·97	−0·20	25·00	24·95	−0·20
15·00	15·01	+0·07	20·00	19·94	−0·30	30·00	30·06	+0·20
20·00	20·01	+0·05	25·00	24·95	−0 20	35·00	34·92	−0·23
40·00	40·00	+0·05	30·00	29·94	−0·20	40·00	39·92	−0·20

RESULTS AND DISCUSSION

The 1 : 2 stoichiometry observed for the oxidation of amino-acids by CAT can be represented as:

$$\mathrm{R''CH\,(NH_2)\,COOH + 2\,RNCl\,Na} \longrightarrow$$
$$\mathrm{2\,RNH_2 + R''CN + CO_2 + NaCl}$$

where $R = p\text{-}CH_3C_6H_4SO_2$ and R'' is $(CH_3)_2CH$ for valine, $(CH_3)_2CHCH_2$ for leucine and $C_6H_5CH_2$ for phenyl alanine.

Paper chromatography[2] was used to identify the sulphonamide ($R_f = 0\cdot905$). Benzyl alcohol saturated with water was used as the solvent with 0·5% vanillin in 1% HCl solution in ethanol as the spray reagent. The nitriles formed, namely, 2-methyl propionitrile, 2-methyl butyronitrile and phenyl acetonitrile were detected by their colour reactions[5] with hydroxylamine and ferric chloride.

The rate of oxidation of the amino-acids by CAT is highly retarded in sulphuric and perchloric acid media. The same trend was observed in the oxidation of unsaturated alcohols by CAT[6,7]. This could probably be attributed to the combined specific inhibitory effect of H^+, SO_4^{--} and H^+, ClO_4^- ions. In contrast, presence of hydrochloric acid accelerated the reaction between CAT and unsaturated alcohols, while a retardation effect is noticed in the present case. It is likely that protonated HOCl is not the species[8] responsible for the oxidation of amino-acids.

The presence of foreign ions such as Ba^{2+}, Zn^{2+}, NO_3^-, $SO_4^=$ and PO_4^{2-} and sodium chloride (up to 0·2 mole) had no effect on the rate or stoichiometry of oxidation of amino-acids by CAT. Further, the stoichiometry is unaffected by a reversal of the order of addition of oxidant and the amino-acid.

The rapid rate of oxidation at pH 4–6 can probably be attributed to the rapid disproportionation of monochloramine–T present in acidified CAT solutions to dichloramine–T and *p*-toluene sulphonamide in this pH range, as suggested by Higuchi *et al.*[9]. It is also to be noted that the isoelectric points of these amino-acids lie within this pH range.

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1. Gowda, N. M. M., Murthy, A. S. A. and Mahadevappa D. S., *Curr. Sci.*, 1975, **44**, 5.
2. Mahadevappa, D. S. and Gowda, N. M. M., *Talanta*, 1975, **22**, 771.
3. Vogel, A. I., *Quantitative Organic Analysis*, Longmann and Greens, London, 1958, Part 3, p. 708.
4. Morris, J. C., Salazar, J. A. and Wineman, M. A., *J. Am. Chem. Soc.*, 1948, **70**, 2036.
5. Soloway, S. and Lipschietz, A., *Anal. Chem.*, 1952, **24**, 898.
6. Mahadevappa, D. S. and Naidu, H. M. K., *Talanta*, 1973, **20**, 349; *Indian J. Chem.*, 1975, **13**, 976.
7. Naidu, H. M. K. and Mahadevappa, D. S., *Curr. Sci.*, 1976, **45**, 216.
8. Mahadevappa, D. S. and Naidu, H. M. K., *Australian J. Chem.*, 1974, 26, 1203; 1975, **28**, 899.
9. Higuchi, T., Ikeda, K. and Hussain, A., *J. Chem. Soc.*, *B*, 1967, p. 546.

ISOLATION AND CHARACTERIZATION OF SOME NEW FORMATE UTILIZING BACTERIA

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ABSTRACT

Five isolates degrading and assimilating formate were isolated from chicken dung. Characterization indicated two different types. One of these belonged to the genus *Alcaligenes* and assimilated formate autotrophically. The other four isolates were identical, belonged to the genus *Protaminobacter* and assimilated formate heterotrophically by the serine pathway.

INTRODUCTION

FORMATE, the simplest of organic compounds is oxidized by a number of bacteria, plants and animals[1–2]. However, only a few bacteria utilize it as a sole carbon source and its assimilation involves two different pathways. Organisms like *Pseudomonas* AM1 assimilate it by the serine pathway[3], while *Ps. oxalaticus*, *Bacterium formoxidans*, *Hydrogenomonas eutropha* Z-1 and *Thiobacillus novellus* (see discussion) assimilate it autotrophically. The fact that only a few cases of autotrophic growth on formate have been reported indicates that this may be a rare type of metabolism as considerations of economy may favour the selection of organisms such as *Ps.* AM1 which conserve the reduction level of formate[4]. The only other C_1 organic compound on which autotrophic growth has been recorded is on methanol by *Micrococcus denitrificans*[5]. As these C_1 compounds are at the borderline between organic and inorganic, a wider study of autotrophic growth on such compounds in a number of genera can be expected to throw light on the biochemical basis of facultative autotrophy.

We report here the isolation and characterization of an autotrophic formate-utilizing bacterium *Alcaligenes* FOR_1 and heterotrophic *Protaminobacter* FOR_2 from chicken dung by enrichment culture with formate as sole carbon source. These isolates differ physiologically from other formate utilizers already known.

METHODS

Isolation and identification.—The media, isolation and identification procedures were the same as described by Chandra and Shethna[6], except that 0·2% (w/v) of sodium formate was substituted for oxalate.

The substrate assimilation and generation time determinations were also done essentially as reported in the above paper.

Preparation of extracts.—The cells were grown as described in Table III, washed and stored below 0° C until used. Extracts in 0·05 M-potassium phosphate buffer pH 7·0 were obtained either by grinding with glass powder or by disrupting in a Raytheon Model DF 101 sonicator for 10 min at 0° C and centrifuged in Sorvall RC2-B at 27000 × g for 15 min. The supernatant was used for the enzyme assays.

Enzyme assays.—All spectrophotometric assays were carried out in a Pye-Unicam spectrophotometer. Formate dehydrogenase (EC 1.2.1.2), hydroxypyruvate reductase (EC 1.1.1.29) and ribulose diphosphate carboxylase (EC 4.1.1.39) were assayed by standard procedures[7,3,8]. Protein was determined by the method of Lowry *et al*[9]. All the cofactors and fine chemicals were obtained from Sigma Chemical Co., U.S.A.

RESULTS AND DISCUSSION

Enrichment yielded several cultures many of which were pink pigmented. Five colourless colonies were studied for the cellular, colony and biochemical characteristics (Table I). Four were identical in their cultural and biochemical properties and were designated as *Protaminobacter* FOR_2 and one was designated *Alcaligenes* FOR_1. The growth on C_1 and C_2 compounds, uptake of oxygen and enzyme studies were determined with one isolate from each type mentioned above.

All the five isolates were negative for the methyl red, Voges-Proskauer, H_2S production, indole formation, cellulose degradation, alginate utilization, and hydrolysis of arginine, casein or gelatin. No growth was observed with 0·2% formate or 0·2% formate *plus* 0·2% acetate, malate or succinate anaerobically. Acetate, malate and succinate supported growth aerobically as the sole carbon source. All were positive for the oxidase, catalase and urea hydrolysis tests. Characteristics in which the isolates varied are shown in Table I.

TABLE I

Description of the isolates

	Alcaligenes FOR_1	*Protaminobacter* FOR_2
Cellular morphology	0·75–1·5 μm, Gram negative coccobacilli, non-motile non-flagellated.	0·75–1·5 μm, Gram negative coccobacilli. Few cells with a central unstained area. Non-motile non-flagellated.
Colonies on formate, gelatin, starch and nutrient agar in 3–4 days	2–3 mm, whitish, translucent, convex, entire-edged smooth, glistening.	2–3 mm, creamish white, opaque, convex, entire-edged, smooth, glistening mucoid.
Growth in nutrient broth (72 hr)	Turbid	Highly turbid.
Tests:		
Action on milk	Acidic	Alkaline
Nitrate reduction*	+	+
Citrate utilization	+	−
Starch hydrolysis	−	+
Acid formation in Hugh and Leifsons' basal medium[26] *with*:**		
Glucose	+	−
Sucrose	−	−
Lactose	+	−
Glycerol	−	+
Fructose	−	+
Arabinose	+	+
Xylose	+	−
Galactose	+	+
Maltose	−	−
Mannitol	−	+

− = No growth or negative reaction;
+ = Growth or positive reaction.
* Nitrate is reduced beyond nitrite in *Pr*.FOR_2.
** No gas was produced with any of the sugars. No acid was produced with glucose, sucrose or lactose anaerobically. *A*.FOR_1 formed acid from lactose aerobically only after 2–4 days.

Even with a heavy inoculum from nutrient agar slants *A*.FOR_1 was noted to alkalise formate only after a 4–5 day period, but subsequent transfers in formate medium led to a more rapid growth. The generation time on 0·3% and 0·5% formate was 13–14 hours with an increasing lag period of 14 and 18 hours respectively as noted even in *Ps. oxalaticus*[10]. Growth on 0·3% formate could not be stimulated by biotin (150 μg/100 ml). One per cent formate completely inhibited growth.

A.FOR_1 assimilated only formate and none of the other C_1 compounds (Table II). *Pr*.FOR_2 assimilated a number of C_1 compounds in addition to formate. The stimulation of growth of *A*.FOR_1 and *Pr*.FOR_2 by yeast extract on oxalate and a few C_1 compounds was also tested as yeast extract was found to 'stimulate growth of *T. novellus* on methanol and formamide (Chandra and Shethna, unpublished observation). However, no increase in growth in presence of yeast extract and C_1 compound compared to controls with yeast extract alone or C_1 substrate alone was noted.

TABLE II

Assimilation of C_1, C_2 and other compounds.

The absorbance was measured at 650 nm in a Bausch and Lomb Spectronic-20 colorimeter after 5–7 days of incubation on a rotary shaker. The turbidity has been recorded without subtracting the control values

Substrates (0·2% w/v)	*Alcaligenes* FOR_1		*Protaminobacter* FOR_2	
	Without yeast extract	With 0·02% yeast extract	Without yeast extract	With 0·02% yeast extract
None	0·05	0·10	0·04	0·09
Formate	0·23	—	0·23	—
Oxalate	0·03	0·11	0·02	0·08
Methanol	0·05	0·11	0·02	0·10
Ethanol	0·04	0·12	0·18	—
Methylamine	0·05	0·09	0·32	—
Formamide	0·06	0·11	0·29	—
Ethylamine	0·06	—	0·04	—
Formaldehyde	0·04	—	0·04	—
Trimethylamine	—	—	0·18	—
Ethanolamine	—	—	0·04	—
Butylamine	—	—	0·03	—
H_2–CO_2–air*	No growth	—	No growth	—

— = Not tested.

* Tested as described by Chandra and Shethna[6].

The biochemical characteristics of *Pr.* FOR_2 indicate that it is a facultative methylotroph closely resembling *Ps. aminovorans*[13] and bacterium 5 B1[16]. On account of its non-flagellation and nutritional characteristics, it has been identified as belonging to the genus *Protaminobacter*. Although it is non-pigmented, it differs from the 4 varieties of *Pr. alboflavus* mentioned in Bergey's Manual[17] in a few nutritional characteristics.

A. FOR_1 possesses cellular, biochemical and nutritional characteristics that agree with the description of the *Achromobacter* genus in Bergey's Manual[17]. However, its properties like facultative autotrophic nature (discussed later), non-flagellation, and acid production from sugars aerobically agree even better with the emended description of the genus *Alcaligenes* by Henrie *et al.*[18]. In view of the uncertain status of the genus *Achromobacter* pointed out by the above authors, we have identified this isolate as belonging to the genus *Alcaligenes*. Its closest resemblance is with *Bacterium formoxidans*[19] but differs from it in the production of acid from milk and sugars, and in not utilizing ethyl alcohol. It is different from *Bacterium formicum*[20] which is known to grow on formate *plus* succinate or malate anaerobically.

The oxidation of formate (Table III) was completely inhibited by 1 mM NaH_2PO_2, an inhibitor of formate dehydrogenase[11] in the case of *A.* FOR_1 similar to the observation with *Ps. oxalaticus*[12]. Only partial inhibition was noted with *Pr.* FOR_2 as in *Ps.* AM1[13]. The oxidation of inorganic substrates, hydrogen and thiosulfate was noted with formate grown *H. eutropha* Z–1[14] and *T. novellus* (Chandra and Shethna, unpublished observation) respectively. However, *A.* FOR_1 and *Pr.* FOR_2 did not oxidize thiosulfate or nitrite.

TABLE III

Oxidative properties of A. FOR_1 and Pr. FOR_2.

Cells were grown in 0·3% formate for 3-4 days with addition of 0·3% formate further after 2 days. The pH was maintained at 7·2 by the addition of 2N HCl. The assay system was the same as described by Chandra and Shethna[6]

Organism	QO_2–μ liters of O_2 uptake/hr/mg dry wt with 30 μ moles formate	% inhibition of QO_2* with 1mM NaH_2PO_2
Alcaligenes FOR_1	38–42 (2)	100 (2)
Protaminobacter FOR_2	31–47 (2)	100 (2)

* Cells were preincubated with NaH_2PO_2 for 15 min. In *Pr.* FOR_2 100% inhibition was observed only in the first 20 min after which period there was only 73% inhibition. In *A.* FOR_1 there was 100% inhibition even after 20 min.

The number in parenthesis indicates the number of determinations.

The specific activity of NAD–linked formate dehydrogenase in *A.* FOR_1 and *Pr.* FOR_2 was 0·260 and 0·182 units/mg protein respectively. The specific activity of hydroxypyruvate reductase in *A.* FOR_1 was low with a value of 0·037 units/mg protein with NADH and 0·02 with NADPH as electron donors. This observation ruled out the heterotrophic serine pathway as the major route of formate carbon assimilation in this culture. Ribulose diphosphate carboxylase the key enzyme of the alternative autotrophic pathway of formate carbon assimilation was investigated in

A. FOR_1 extract and was detected at a specific activity of 10 m. moles of phosphoglyceric acid formed/min/mg protein. It is reported[27] that in some autotrophic bacteria natural macromolecular inhibitors of ribulose diphosphate carboxylase occur giving rise to low activities of this enzyme. It is possible that this could be the reason for the low activity of ribulose diphosphate carboxylase in *A.* FOR_1 and needs further investigation. It is therefore tentatively concluded that *A.* FOR_1 grows autotrophically on formate. In *Pr.* FOR_2 the specific activity of hydroxypyruvate reductase with NADH as electron donor was high (0·40 units/mg protien) indicating the heterotrophic serine pathway as the major metabolic route of formate carbon assimilation as in *Ps.* AM1 and AM2[15]. In view of the good activity of this enzyme the presence of ribulose diphosphate carboxylase was not further examined.

Relatively few attempts have been made to isolate formate utilizing bacteria[19,21,22]. The autotrophic pathway of formate assimilation has been conclusively demonstrated only in 4 non-photosynthetic organisms, *viz., Ps. oxalaticus*[8], *H. eutropha* Z–1[14], *B. formoxidans*[19] and *T. novellus*[23] by radioactive and/or enzymic studies. Evidence in *Nitrobacter winogradskyi* and *Thiobacillus* A2 is lacking although the authors have reported that they utilize formate[24–25]. Of the four organisms mentioned, *B. formoxidans* is the only autotroph obtained from soil directly by enrichment with formate. Our results indicate that *A.* FOR_1 obtained by similar procedure is one of the few organisms that can assimilate formate autotrophically and *Pr.* FOR_2 is a heterotroph like most facultative methylotrophs.

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1. Quayle, J. R., *Adv. Microb. Physiol.*, 1972, 7, 119.
2. Chandra, T. S. and Shethna, Y. I., *Ant. van Leeuwenhoek.*, 1975, **41**, 465.
3. Large, P. J. and Quayle, J. R., *Biochem. J.*, 1963, **87**, 386.
4. Storm, T., Ferenci, T. and Quayle, J. R., *Ibid.*, 1974, **144**, 465.
5. Cox, R. B. and Quayle, J. R., *Ibid.*, 1975, **150**, 569.
6. Chandra, T. S. and Shethna, Y. I., *Ant. van Leeuwenhoek.*, 1975, **41**, 101.
7. Johnson, P. A., Jones-Mortimer, M. C. and Quayle, J. R., *Biocheim. Biophys. Acta*, 1964, **89**, 351.
8. Blackmore, M. A. and Quayle, J. R., *Biochem. J.*, 1968, **107**, 705.
9. Lowry, O. H., Rosebrough, N. J., Farr, A. L. and Randall, R. J., *J. Biol. Chem.*, 1951, **193**, 265.
10. Dijkhuizen, L. and Harder, W., *Ant. van Leeuwenhoek.*, 1975, **41**, 135.
11. Takamiya, A., *J. Biochem.* (Tokyo), 1953, **40**, 415.
12. Quayle, J. R. and Keech, D. B., *Biochem. J.*, 1959, **72**, 623.
13. Peel, D. and Quayle, J. R., *Ibid.*, 1961, **81**, 465.
14. Namsaraev, B. B., Nozhevnikova, A. N. and Zavarzin, G. A., *Mikrobiologiya* (Transl.), 1971, **40**, 675.
15. Blackmore, M. A. and Quayle, J. R., *Biochem. J.*, 1970, **118**, 53.
16. Colby, J. and Zatman, L. J., *Ibid.*, 1973, **132**, 101.
17. *Bergey's Manual of Determinative Bacteriology*, By Breed, R. S., Murray, E. G. D. and Smith, N. R. The Williams and Wilkins Company, Seventh Edition, 1957.
18. Henrie, Margaret, S., Holding, A. J. and Shewan, J. M., *Int. J. Syst. Bact.*, 1974, **24**, 534.
19. Sorokin, Yu. I., *Mikrobiologiya* (Transl.), 1961, **30**, 337.
20. Shaposhnikov, V. N. and Loriya, Zh. K., *Dokl. Acad. Nauk., SSSR*, 1967, **175**, 721.
21. Bhat, J. V., *J. Ind. Inst. Sci.*, 1956, **38**, 43.
22. Pavri, Khorshed M. and Bhat, J. V., *Proc. Ind. Acad. Sci.*, 1953, **38 B**, 171.
23. Shethna, Y. I., *Ind. J. Biochem.*, 1967, **4**, 34.
24. Van Gool, A. and Laudelout, H., *Biochim. Biophys. Acta*, 1966, **127**, 295.
25. Taylor, B. F. and Hoare, D. S., *J. Bacteriol.*, 1969, **100**, 487.
26. Hugh, R. and Leifson, E., *Ibid.*, 1953, **66**, 24.
27. McFadden, B. A., *Bacteriol. Rev.*, 1973, **37**, 289.

NEW EVIDENCE OF MAGNETIC POLE VARIATIONS

Scientists, including a team from the Australian National University in Canberra, are uncovering increasing evidence that the earth's magnetic poles have migrated dramatically in the past. The Australian National University's Research School of Earth Sciences research team has developed techniques which may allow them to reconstruct in detail the variations of the geomagnetic field in Australia over the past 30,000 years. The technique involves taking long, undisturbed oriented cores of sediment from the bottom of lakes and analysing their magnetic records. (Australian Information Service, Canberra, ACT 2600.)

LETTERS TO THE EDITOR

SPIN OF THE 1251 KeV LEVEL IN ^{97}Nb

THE ground state spin of ^{97}Zr is estimated[1] to be 1/2+. The 1251 KeV level of ^{97}Nb, first proposed by Hofstetter and Sugihara[2], is fed by the 1410 KeV beta group from ^{97}Zr with an intensity of 3·1% and a log ft value 8·1. From different measurements[2,3,4] the possible spin-parity characters of the 1251 KeV level are $1/2^-$, 3/2+ and 5/2+. The corresponding 1·41 MeV beta transition, then, shall be of non-unique or unique first forbidden type or a second forbidden case. A measurement of β–γ angular correlation will shed light on the spin-parity nature of the 1251 KeV level. Hence a careful measurement of the anisotropy of the 1·41 (β)-0·508 MeV (γ) cascade in the decay of ^{97}Zr nucleus is undertaken. Such an attempt does not appear to have been done earlier. The experiment is carried out on a conventional fast-slow scintillation assembly.

The value of β–γ anisotropy, A, thus obtained is 0·481 ± 0·124. From this, the value of the correlation coefficient, A_2 (β, γ) = ϵ, is determined to be 0·297 ± 0·086.

The large value of A excludes the possibility of the spin of the 1251 KeV level being $1/2^-$ inasmuch as, A is zero for an intermediate state with a spin 1/2. If it is 3/2+, the beta transition will be an allowed one. The value of A is zero in this case also, which is contrary to the experiment. Thus the 3/2+ possibility is excluded. A 5/2+ assignment[4] classifies the transition as second forbidden type. However, the log ft value 8·1 is too low for the beta transition to be of second forbidden type. Also, the absence of a gamma transition from 1251 KeV state to the ground state of ^{97}Nb, which will be of E 2 type if the spin of the initial state were 5/2+, rules out this possibility. Finally we have to consider the two choices $3/2^-$ and $5/2^-$. The latter possibility classifies the 1·41 MeV beta transition to be of unique first forbidden type for which A_2 (β) could be theoretically computed[5] to be – 0·75. A_2 (β) determined from experimental correlation coefficient A_2 (β, γ) is – 0·556 ± 0·160. From the close agreement between these two within experimental uncertainties, we infer the spin of the 1251 KeV level as $5/2^-$. On the other hand, if the spin of the 1251 KeV level were to be $3/2^-$, the 1410 KeV beta transition would be a non-unique first forbidden transition. The ξ of the transition is 13·14, which is greater than $W_0 - 1$ (= 2·76). So if the ξ approximation were to be valid in this case, we expect isotropic β–γ directional correlation and a statistical shape. A measurement of the spectrum shape of the 1·41 MeV transition would help not only in confirming or rejecting the $3/2^-$ possibility but also in assigning to the 1251 KeV level of ^{97}Nb uniquely $5/2^-$ or $3/2^-$ character, which are concluded in the present investigation, though the $5/2^-$ assignment seems more probable.

The Laboratories for
Nuclear Research,
Andhra University,
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1. Cohen, B. L., *Phys. Rev.*, 1961, 125, 1358.
2. Hofstetter, K. H. and Sugihara, T. T., *Nucl. Phys.*, 1970, 140 A, 658.
3. Megli, D. G., Agin, G. P., Potny, V. R. and Mandeville, C. E., *Z. Physik.*, 1970, 234, 95.
4. Murty, D. S., Jagan, P. and Lakshminarayana, V., *Can. J. Phys.*, 1971, 49, (12) 1649.
5. Steffen, R. M., *Alpha, Beta and Gamma Ray Spectroscopy*, 1965, Vol. 2, Ed. By K. Siegbahn.

ATOMIC ABSORPTION SPECTROPHOTOMETRIC ANALYSIS OF THE SEEDS OF *STRYCHONUS POTATORAM* LINN.

Strychonus potatoram Linn. is a natural polyelectrolyte of local origin and belongs to the family "STRYCHNACEAE" (formerly Loganiaceae). The seed is used for clearing muddy water[5] and has long been recognised for its medicinal properties[1,2]. Complex polysaccharides and other organic constituents[3,4] have been reported in these seeds.

In the present studies the ash of the seeds was used for the estimation of metallic constituents by standard methods. However, metals like Ni, Co, Bi, Ag, Zn and Fe were estimated by atomic absorption spectrophotometric analysis (Perkin Elmer Make A.A.S.).

The ash was mixed with 40 ml of HCl (1 : 1) and digested for 30 minutes on a water-bath. The extract was used for the qualitative and quantitative analysis of inorganic constituents of the seeds. Elements were detected by semi-micro and micro analysis. Blank tests were also carried out for comparison.

The following are the results of analysis expressed in percentages of the seed:

(1) Oorganic matter: 78·7%, (2) Silica: 0·0155%. (3) Ca: 0·225%, (4) Mg: 0·131% and (5) Mn: 0·0013%. The atomic absorption spectrophotometric analysis was carried out for nickel, cobalt, bismuth, silver, zinc and iron and the results are as follows in ppm:

Ni: 220, CO: 30, Bi: 50, Ag: 15, Zn: 190 and Fe: 120.

Generally, silver is not obtainable even in trace amount in any Indian plant[6]. Deb *et al.*[6] have reported 5 ppm silver in edible algae of Manipur. The present studies, however, indicate the presence of 15 ppm silver in Nirmali Seeds. The presence of 220 ppm of nickel seems to be quite high.

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Institute of Science, V. S. DARSHANE.
Nagpur, *March* 12, 1976. V. V. DESHPANDE.

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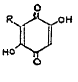
1. Kirtikar, K. R. and Basu, B. D., *Indian Medicinal Plants*, Lalit Mohan Basu, Publishers, Allahabad, 1935, 3, 1647.
2. Chopra, R. N., Nayar, S. N. and Chopra, I. C., *Glossary of Indian Medicinal Plants*, C.S.I.R., New Delhi, 1956, p. 236.
3. Haksar, C. N. and Khendurkar, H. G., *J. Vikram. UNI*, 1960, 1, 24.
4. Alok Singh, and Bajpai, R. K., *J. Indian Chem. Soc.*, 1975, 52, 768.
5. Bulusu, K. R. and Sen, *Environmental Health*, 1962, 4, 233.
6. Deb, D. B. *et al.*, *Curr. Sci.*, 1974, 43, 629.

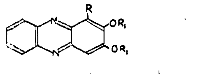
SYNTHESIS OF A NEW *PHENAZINE*

PHENAZINE systems are well known for their utility as dyes. Several studies have been made to establish their pharmacological utility. *Streptomyces griseoluteus* is known to produce phenazine compounds having carboxyl group or alkyl group[1]. Hydroxy, methoxy and aminophenazines have been thoroughly studied for their insecticidal value[2]. It is therefore proposed to synthesize a hydroxyphenazine with an alkayl side chain, which may have useful physiological action.

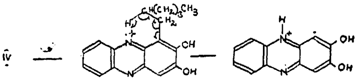
2, 5-Dihydroxy-1 : 4-benzoquinone [I] reacts with orthophenylenediamine forming 2, 3-dihydroxyphenazine [II][3]. Embelin, 2, 5-dihydroxy-6-undecyl-1 : 4-benzoquinone [III] has been condensed with orthophenylenediamine in 1 : 1 molar proportion in acetic acid as solvent. The reaction mixture on working up gave a red crystalline substance m.p. 225° C. It gave a positive ferric reaction and it developed a yellow coloration in aqueous sodium-hydroxide. Mass spectrum indicated the M+ peak (366) which is in agreement with the molecular formula $C_{23}H_{30}O_2N_2$. I.R. spectra data 3200 cm^{-1} (—OH), 1625 cm^{-1} (—C=N—) and uv absorptions at 253·5 nm (log ε, 5·24), 260 nm (log ε, 5·24) 391 nm (log ε, 4·84), showed the compound to be a phenazine derivative.



I R=H
III R= $-(CH_2)_{10}CH_3$



II R = R_1 = H
IV R = $-(CH_2)_{10}CH_3$, R_1 = H
V R = $-(CH_2)_{10}CH_3$, R_1 = $-COCH_3$



IV

VI

The presence of hydroxyl groups has been evidenced by the formation of acetate [V], m.p. 118° C, by refluxing the compound with acetic anhydride-sodium acetate. [V] showed the uv absorptions 253·5 nm (log ε, 5·13) and 365 nm (log ε, 4·22) characteristic of phenazine derivatives Proton-magnetic resonance spectrum recorded on Brucker WH 90 in deuterodimethyl sulfoxide as solvent showed 7·23 δ (H, aromatic) 7·72 δ (2 H, aromatic) and 8·02 δ (2 H, aromatic) which is in agreement with the structure [IV]. Protons of the aliphatic side chain were observed in the region 2·5 δ to 3·5 δ. prominent peaks recorded in the mass spectrum (70 ev) 366 (M+, 5·8%); 226 (M+ — 140, 12·5%); 212 (M+ — 154, 100%) and 184 (212–28, 90·5%), confirm the structure [IV] for the condensation product of embelin with orthophenylenediamine. Intense peak at m/e 212 is attributed to the formation of radical ion [VI]. Aliphatic side chain fragment $C_{11}H_{22}^{(+)}$ (154) has also been recorded.

More conclusive evidence for the structure of the condensation product [IV] has been obtained through the NMR spectrum of the acetate [V] in deuterochloroform, which showed signals 0·95 δ (3 H, *t*, aliphatic CH_3); 1·36 δ (18 H aliphatic protons); 3·45 δ (2 H, *t*, benzylic protons); 2·47 δ (3 H, *s*, $-COCH_3$); 2·52 δ (3 H, *s*, $-COCH_3$); and 8·13 δ (1 H, *s*, aromatic H). Four aromatic protons were indicated by signals at 7·92 δ and 8·31 δ.

Further work in the field is in progress. Author is thankful to Prof. B. Franck for N.M.R., I.R.

and mass spectra and to Sri. P. S. Sastry to uv spectra. Thanks are due to Prof. E. V. Sundaram, Head of the Department of Chemistry and to Prof. K. Venkata Ramiah, Director, P.G. Centre, for their encouragement.

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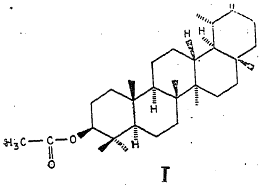
1. Koko Yagishita, *J. Antibiotics* (Japan), 1960, 17 A, 8396.
2. Smith, L. E., *C.A.*, 1942, 36, 2644 ; *Ind Eng. Chem.*, 1942, 34, 499.
3. Kehrmann, *Ber.*, 1890, 23, 905 ; Bulock, *J.C.S.*, 1955, p. 575.

CONVERSION OF TARAXASTERYL ACETATE INTO TARAXASTANE 3β-20 DIOL

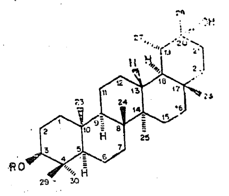
EPOXIDATION (PBA) of taraxasteryl acetate[1] (I) gives a mixture of two stereoisomeric epoxides (TLC) (C_{20}-epimers). One of these epoxides (less polar), $C_{32}H_{52}O_3$, M+ 484, m.p. 236–38° (hexane + 10% benzene) has been isolated in the TLC pure state by chromatography. It shows IR absorptions at 1724, 1250 cm^{-1} (acetate) and NMR (CCl_4) signals at τ : 9·17, 9·14, 9·07, 9·04, 8·97, 8·9 (21 H, methyls at C_4, C_8, C_{10}, C_{14}, C_{17} and C_{19}) ; 8·07 (3 H, *s*, acetate methyl at C_3) ; 7·4 (2 H, *s*, CH_2 protons at C_{28}) and 5·6 (1 H, *t*, C_3-proton).

LAH reduction of the epoxide gives a diol (II), $C_{30}H_{52}O_2$, M+ 444, m.p. 266–68°, $(\alpha)_D$ − 9° (*c*, 1·5). It shows IR absorptions at 3344 (OH) and NMR ($CHCl_3$) signals at τ : 9·2, 9·14, 9·0, 8·93, 8·9 (21 H, methyls at C_4, C_8, C_{10}, C_{14}, C_{17} and C_{19}) ; 8·8 (3 H, *s*, methyl at C_{20}) and a triplet at 6·8 (1 H, C_3-proton). Acetylation of (II) (Ac_2 O/Py) affords a monoacetate (III), $C_{34}H_{54}O_3$, M+ 486, m.p. 276–80° (hexane), $(\alpha)_D$ ± 0°. It shows IR absorptions at 3448 (OH), 1718, 1266 cm^{-1} (acetate) and NMR (CCl_4) signals at τ : 9·18, 9·15, 9·0, 8·9 (21 H, methyls at C_4, C_8, C_{10}, C_{14}, C_{17} and C_{19}) ; 8·86 (3 H, *s*, methyl at C_{20}) ; 8·0 (3 H, *s*, acetate methyl at C_3) and a triplet centred at 5·57 (1 H, C_3-proton).

Two taraxastane 3 β-20 diols which are epimeric at C_{20} have been described in literature[2,3]. A comparison of the IR spectrum of diol (II) with that of 20-epi-ψ-taraxastane, 3 β-20 diol[3], isolated from black dammar resin, suggests a close relationship between the two compounds. However, some differences were observed in the NMR spectra of their monoacetates[3] especially in the methyl region signals. In addition, the physical constants especially, the optical rotation of the two diols and their monoacetates are found to be different [Lit.[3] records for 20-epi-ψ-taraxastane-3 β-20 diol, m.p. 261–63°, $(\alpha)_D$ ± 0° ; monoacetate, m.p. 266–67°, $(\alpha)_D$ + 23°].



I



II. R=—H
III. R=—COCH₃

However, the physical constants of both (II) and (III) are in close agreement with those reported for ψ-taraxastane 3 β-20 diol[2] and its monoacetate. [Lit.[2] records for ψ-taraxastane 3 β-20 diol, m.p. 270–72°, $(\alpha)_D$ − 10·9°, monoacetate, m.p. 281–84°, $(\alpha)_D$ − 1·5°].

It therefore appears that diol (II) is identical with ψ-taraxastane 3 β-20 diol, isolated from manila elemi resin. A direct comparison between the two, however, could not be made due to the non-availability of the sample of ψ-taraxastane 3 β-20 diol.

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1. Namboodiripad, C. P., Kulkarni, G. H. and Kelkar, G. R., *Curr. Sci.*, 1968, 37, 550.
2. Morice, M. and Simpson, C. E., *J. Chem. Soc.*, 1940, p. 795 ; *Ibid.*, 1941, p. 181.
3. Hinge, V. K., Paknikar, S. K., Das, K. G., Bose A. K. and Bhattacharyya, S. C., *Tetrahedron*, 1966, 22, 2861.

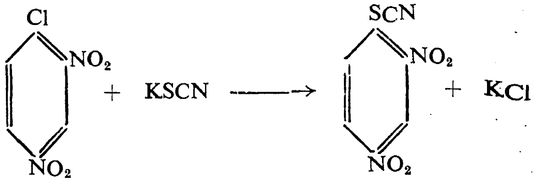
STUDIES ON PESTICIDES

Part I. Some Halogeno-polynitro Phenyl, Tolyl and Naphthyl Thiocyanates

NITRO phenyl, thiocyanates have been claimed to be toxicants for fungi[1,2], bacteria, moulds and other pests[3,4]. The pesticides also show some tuberculostatic and acaricidal activity[5]. Incorporation of one or more substituents of elecrophilic character in the ortho or para positions to the thiocyanate group results in an increase of both bacteriostatic and fungistatic activity[5]. It has been suggested that these compounds exert their anti-microbial activity by an intra-cellular interaction of —SH enzyme[5].

In view of the known pesticidal activity of nitro phenyl thiocyanates, the synthesis of some analogues of these compounds containing methyl, halogens and other substituents at various positions in the ring and their pesticidal activity have been undertaken.

These compounds have been prepared[6] by heating halogenonitrobenzene, toluene or naphthalene (0·01 M) with potassium thiocyanate (0·01 m) in methanolic solutions (20–40 ml) till the product separated. The compound so obtained has been crystallised from anhydrous ethanol. These are yellow crystalline compounds insoluble in water but soluble in organic solvents. Their melting points, yield, etc., are given Table I. The compounds have been analysed for C, N, H and S and the analytical results agreed with the calculated within the experimental errors.



Microbial activity.—The compounds have been tested for their fungicidal activity against *Alternaria solani* and *Aspergillus niger* by poisoned food technique. The fungus is grown on potato-dextrose agar-agar media containing various concentrations of the test compound. A concentration at zero served as check. After seven days of the inocula-

TABLE I

$R-S-C\equiv N$

Halogeno nitro benzene used	R	M.P.* °C	Yield %	*Aspergillus niger* 100	50	25	10	0	*Alternaria solani* 100	50	25	10	0
				Concentration in ppm — Radial growth of the fungus colony in millimeters									
1-Chloro-2:4-DN	2:4-DNP	138	70	nil	14	14	22	64	nil	nil	32	52	56
1-Chloro-2:6-DN	2:6-DNP	71	50	nil	55	70	81	85	nil	nil	nil	20	50
1-Chloro-2:4:6-TN	2:4:6-TNP	100*d*	80	70	83	86	88	90	36	41	46	52	52
1:2-Dichloro-4:6-DN	6-Chloro-2:4-DNP	143	40	..	..	..	..	..	..	..	..	..	..
1:3-Dichloro-4:6-DN	5-Chloro-2:4-DNP	193	50	56	60	62	66	80	45	56	58	60	72
1:4-Dichloro-2:6-DN	4-Chloro-2:6-DNP	102	55	nil	nil	70	75	80	nil	nil	nil	27	46
1-Chloro-4:6-DN-3-methyl	5-methyl-2:4-DNP	70	65	nil	nil	30	44	66	nil	nil	10	42	58
1-Chloro-2:4:6-TN-5-methyl	3-methyl-2:4:6-TNP	139	65	nil	nil	nil	nil	80	nil	nil	nil	nil	46
1:4-Dibromo-2:6-DN	4-Bromo-2:6-DNP	105	50	nil	nil	nil	44	61	nil	60	40	48	58
1-Chloro-2:4-DN-naphthalene	2:4-dinitro-naphthyl	150*d*	50	..	..	..	..	..	..	..	..	..	..

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* All melting points are uncorrected.

N.B.—DN, TN, DNP and TNP refer respectively to dinitro, trinitro, dinitro phenyl and trinitro phenyl.

tion of the fungus the radial growth is measured. The temperature is kept between 24–28° during the whole period. The radial growth of the fungi at various concentrations of the thiocyanates and in the absence of the compound are given in Table I.

It is interesting to note that a methyl group at meta position and nitro group at ortho and para position to the thiocyanato group increases fungicidal activity of the thiocyanate. The 3-methyl 2 : 4 : 6 trinitro-phenyl thiocyanate is found to be the most active towards both the test fungi.

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Saharanpur, *June* 10, 1976.

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1. Bara, I., *Kiserl Kozlemeny, C. Kertesz,* 1960, 53 (2), 79.
2. Sys, S. and Soenene, A., *Agricultura* (Lauvain), 1969, 17 (2), 65.
3. Taseng-sung Wang, Hsi Nan Nung Yeh K'ohsuch, 1958, 1, 73.
4. Flenner, A. L. and Kabert, R. A., *U.S. Pat.*, 1947, Dec. 2, 433 ; 106, *Chem. Abstr.*, 42, 2723 *d.*
5. Zsolnai, I. T., *Arzneimittel-Forsch.*, 1966, 16 (7), 870.
6. Albert, L. F. and Russell, A. K., *U.S. Pat.*, 2, 433, 106, Dec, 23, 1947 ; *CA.*, 1948, 42, 2723 *d.*

TANNINS OF *CAESALPINIA PULCHERRIMA* BARK

The seeds[1,2] of *Caesalpinia pulcherrima* have been reported to contain a galactomannan. The stem bark of this plant is highly astringent and is widely used as an abortifacient and as an emmenagogue. It has now been investigated and found to contain gallic acid, ellagic acid, leucodelphinidin and a new tannin, which has been studied in detail.

Extraction

Following a general procedure for the extraction of polyphenols the bark was extracted with ethanol. The ether soluble fraction from the concentrated ethanolic extract was found to contain gallic acid, ethyl gallate and traces of ellagic acid. Ethyl acetate extracted a leucoanthocyanidin along with a tannin (A). The mother liquor was further concentrated to a viscous residue and macerated with acetone, which extracted some more of the tannin (A). From the residue free ellagic acid could be extracted out with ethanol containing traces of pyridine. The leucoanthocyanidin could be characterised as leucodelphinidin by its characteristic colour reactions and spectral studies. On acid treatment, it could be converted to its corresponding anthocyanidin delphinidin, which was found to be identical with an authentic sample isolated from *Solanum melongena* fruits, in its colour reactions, paper chromatography and λ_{max} (560 mμ, ethanolic HCl).

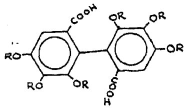
The isolation of ethyl gallate, which is usually isolated as an artefact formed as a result of alcoholysis of depside links present in tannins, during the extraction with ethanol, led us to modify the method of extraction. The bark was extracted with water at room temperature. The combined extract was demineralised over a mixed bed of cation and anion exchange resins to constant conductance and then concentrated under diminished pressure to a syrupy mass. Maceration with ether of the viscous residue gave some gallic acid and further extraction with ethyl acetate gave a mixture of gallic acid, leucodelphinidin and another tannin (B). Maceration with acetone of the remaining sticky residue gave some more amount of tannin (B). The acetone concentrate was charged over a silica gel (deactivated) column and eluted with benzene-acetone mixture. Final crystallisation of the last fractions from acetone-ether mixture gave a colourless semi-crystalline compound, which was found to be a homogeneous entity by paper chromatography and TLC.

Tannins (*A*) and (*B*)

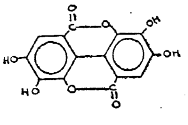
Both these tannins gave positive Molisch test and blue-black precipitate with ferric chloride, suggesting these to be polyphenolic glycoside, but positive colour reaction with aniline hydrogen phthalate reagent[3], confirmed their non-glycosidic nature. Tannin (A) on alkali hydrolysis gave D (+)-glucose, gallic acid and ellagic acid, suggesting thereby that these acids are possibly esterified with the glucose moiety.

Tannin (B) on acid as well as alkali hydrolysis gave glucose, gallic acid and ellagic acid. The quantitative estimation of glucose shows the presence of 19% glucose. Ellagic acid precipitated out almost quantitatively during hydrolysis and could be directly weighed and found to be 30%. Gallic acid was found to be 55% by potentiometric titration. This calculates to 3 moles of gallic acid and one mole of ellagic acid per mole of glucose. The acetate of this tannin analysed for 15 acetyl groups ($-COCH_3$, 40·5%) per mole of acetate. Methylation was done with diazomethane. IR spectrum of the methyl ether confirmed the absence of any free hydroxyl group. The methyl ether on acid as well as alkaline hydrolysis gave three acids, which were identified as trimethyl gallic acid, 3 : 4-dimethyl gallic acid and hexamethoxydiphenic acid (I *b*). These three acids were found to be present in the ratio 2 : 1 : 1 respectively, as indicated by comparative paper chromatography carried out with an artificial mixture of the authentic acid.

As hexamethoxydiphenic acid (I *b*) is one of the products of hydrolysis of the methylated tannin it is assumed that hexahydroxy diphenic acid (I *a*) must be linked to the glucose core through ester linkages involving its two carboxyl groups. Moreover, hydrolysis of the tannin itself gave ellagic acid (II). It is well known that hexahydroxydiphenic acid easily cyclises to form its dilactone ellagic acid under the conditions of the hydrolysis.



(I) (a) R=H, (b) R=CH_3



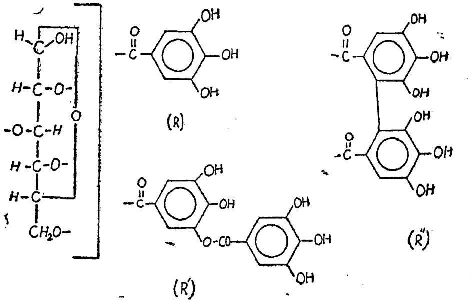
(II)

The proportion of trimethyl gallic acid, dimethyl gallic acid and hexamethoxyldiphenic acid, obtained by hydrolysis of the methylated tannin as 2 : 1 : 1 respectively, shows that one hydroxydiphenic acid, one gallic acid and one *m*-digallic acid are esterified with 2, 3, 4 and 6th carbons of glucose. Two of these hydroxyls may be involved in ester linkages with the two carboxyls of hexahydroxydiphenic acid. These hydroxyls may be 2 and 4, or 4 and 6 or 3 and 6. Out of the remaining two hydroxyls one is esterified with gallic acid and the other with a digalloyl chain.

On controlled methanolysis the tannin (B) gave traces of gallic acid, methyl gallate and another tannin, which compared on paper chromatography and TLC with the one obtained from the ethanolic extract of the bark (Tannin A). This tannin on hydrolysis gave gallic acid, ellagic acid along with glucose. Gallic acid estimation (43%) of this tannin suggested that there may be two gallic acid units per mole of glucose. This suggested that the remaining one gallic acid must be linked with a depside link. As no *m*-digallic acid could be detected during methanolysis, the possibility of a trigalloyl chain is completely excluded.

A number of alternative structures can be assigned to this tannin based on the above data, as it is difficult to assign definite positions to gallic acid, digalloyl and hexahydroxydiphenic acid units on the glucose moiety. Therefore, the distribution of R, R′ and R″ is of course random.

This tannin from *C. pulcherrima* stem bark is special in containing *m*-digalloyl unit in addition to hexahydroxydiphenic acid. A polygalloylated chain has not so far been reported to be present in any of the naturally occurring ellagitannins. Moreover, the presence of both leucodelphinidin and gallotannin in the same source has also some biogenetic significance, as probably this plant specialises in biosynthesising pyrogallol unit.



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---

1. Bains, G. S.. Bhatia, D. S. and Subrahmanyam, V., *J. Indian Chem. Soc., Ind. and New Ed.,* 1956, 19, 103.
2. Morimoto, J. Y., Unran, Ireve, C. J. and Unran, A. M., *J. Agr. Food Chem.,* 1962, 10, 134.
3. Hough, L., Jones, J. K. N. and Wadman, W. H., *J. Chem. Soc.,* 1950, p. 1702.

A NEW SPRAY REAGENT FOR THE IDENTIFICATION AND DETERMINATION OF ORGANO-PHOSPHORUS INSECTICIDES BY THIN LAYER CHROMATOGRAPHY

Various reagents are described in literature for the detection of organo-phosphorus insecticides on thin layer chromatographic plates. Most common ones are palladium (II) chloride[1,5], bromine-fluorescein-silver nitrate[2,3], Congo red[4], mercuric nitrate-diphenylcarbazone[6], mercurous nitrate[7], reduction, diazotisation and coupling with N-1-naphthylethylene-diamine[8], ferric chloride-sulfosalicylic acid[9], etc. To a large extent they are not sensitive and are susceptible to impurities; some are non-specific. Though some of these reagents[1,2,4,8,10], are sensitive with detection limit as low as 1 to 5 mcg., they are only useful for the detection of the compounds in pure form. In forensic toxicology the extraction of these compounds from biological materials is always followed with the co-extraction of other interfering substances including fatty material. Purification of such extract is time consuming and generally involves the loss of the insecticides which may escape detection with the usual reagents. Because of the high toxicity of the organo-phosphorus insecticides, a need was felt for a more specific and sensitive reagent which can detect these compounds even in the presence of fatty material. In this paper a new spray reagent, *viz.*, mercuric nitrate followed by potassium ferrocyanide has been described for their detection and determination on thin-layer chromatograms. This reagent is specific for organo-phosphorus insecticides only ; and organo-chloro insecticides, carbamate insecticides, and fatty material generally extracted along with these compounds do not interfere. The sensitivity of the reagent is ≈ 5 mcg.

Experimental

Different solutions in methanol had concentration of 1 mg/ml of the active ingredient. Quantity of these standard solutions spotted was 5 μg. Each of the reference compounds were spotted on to the Silica Gel G layer (1 mm) previously activated at 110° C for half an hour. The plates were then developed in the solvent system Hexane-Acetone (9+1) by the ascending technique. The plates were air-dried and sprayed with reagent (1) 0·5 gm of mercuric nitrate dissolved in 100 ml of water and (2) 1 gm of potassium ferrocyanide dissolved in 100 ml of water (freshly prepared). Bluish-green spots were observed after keeping the plate for about 5 minutes at room temperature.

Discussion

The R_f values of various organo-phosphorus insecticides are given in Table I. The compounds which are listed above are generally present in

The R values of various organo-phosphorus

TABLE I

Identification of organo-phosphorus insecticides on T.L.C. plates

Sl. No.	Name of Insecticide	R_f. Values		Colour of the spot	Minimum amount detected
1.	Parathion	0·54	P	Green	5 mcg
2.	Parathion-Methyl	0·21	S	Yellow-green	5 mcg
		0·23–0·45	P	Blue–green	
3.	Malathion	0·09	S	Greenish	5 mcg
		0·43	P	Green	
4.	Diazinon	0·06	S	Bluish	5 mcg
		0·4	P	Green	
5.	Rogor (Dimethoate)	0·16	S	Green	5 mcg
		0·42	P	Yellow–greenish	
		0·54	S	Green	
6.	Fenitrothion (Sumithion)	0·43		Green	5 mcg
7.	Fenthion (Baytex, Dalf)	0·28	S	Green	5 mcg
		0·53	P	Green	
8.	Fensulfothion (Dasanit)	0·13	S	Green	5 mcg
		0·63	P	Yellow–green	
9.	Bayer 78418	0·42	P	Blue	50 mcg
		0·85	S	Blue	
10.	DDVP (Dichlorovas)	—	—	Blue	50 mcg
11.	Demacron (Phosphamidon)	—	—	Blue	50 mcg
12.	Metasystox	0·16	P	Yellow	10 mcg
		0·33	P	Yellow	

P = Principal spot;   S = Secondary or faint spot.

ing cases involve the use of such formulations, which are readily available in the market, it was more imperative to study these market products as such on TLC rather than pure compounds. In some cases 3–4 spots have been obtained, this is due to the fact that many times compounds are present in the impure form in a mixture of 2–3 related substances. The intensity of the spots increases as the time passes and the colour remains stable for several days. About 5 mcg of Parathion, Methyl Parathion, Diazinon, Dimethoate, Sumithion, Fenthion, Fensulfothion and Metasytox can be detected, on TLC plates with this reagent, while the sensitivity for DDVP, Phosphamidon, and Bayer 78418 is ≈ 50 mcg. This reagent has been used successfully for the identification and determination of these insecticides in biological, visceral material in poisoning cases.

We are thankful to Dr. B. N. Mattoo, Director, for his keen interest in the work.

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1. Baümler, J. and Rippstein, U. S., *Helv. Chim. Acta*, 1961, **44**, 1162.
2. Walkar, K. C. and Beroza, M., *J. Ass. Off. Agr. Chem.*, 1963, **46**, 250.
3. Devidek, J. U. J. and Pokorny, *Z. Lebensmitt Unters*, 1961, **115**, 113.
4. Irudayaswamy, A. and Natarajan, A. R., *Analyst*, 1965, **90**, 503.
5. Kurt Randerath, *Thin Layer Chromatography*, Academic Press, New York and London, 1965, p. 205.
6. Joglekar, V. D. and Mahal, H. S., *Archivfur, Kriminologie*, 1968, **142**, 171.
7. Kawale, G. B., Joglekar, V. D., Barve, V. P. and Mahal, H. S., *Science and Culture*, 1972, **38**, 373.
8. Erne, E., *Acta Pharma, Tox Kbh.*, 1958, **14** (2), 173.
9. Vutreshni Bolesti, Untrechni Bolesti, Nauchno, *Druzhestro, Internistite*, 1964, **3** (4), 573.
10. Kovacs, M. F., *J. Ass. Off. Agr. Chem.*, 1964, **47**, 1097.

ZINC (II), CADMIUM (II) AND MERCURY (II) COMPLEXES OF *BIS*-(VANILLYDENE) ETHYLENEDIAMINE

Introduction

IN our previous papers[1,2] on vanillydene-aniline complexes with bivalent metal ions, we have shown that they form 1 : 1 complexes. The Zn (II), Cd (II) and Hg (II) have been given the chloro-bridged structures in which they all exhibit co-ordination number four. Vanillydene-*o*-toluidine differs from vanillydene-aniline in a manner in which the OH and OCH_3 groups are involved in the coordination rather than the $C{=}N$. Thus it shows neutral bidentate behaviour. To have departure from these ligands we have used *bis* (vanillydene)-ethylenediamine which contains two $C{=}N$ groups, two OCH_3 groups and two OH groups. The geometry of the ligand has aroused our interest to prepare the complexes of it. It is interesting to know whether the ligand shows bidentate behaviour coordinating through azomethine groups or quadridentate behaviour involving both OH and OCH_3 groups which are ortho to each other on the aldehyde residue.

Experimental

The chemicals used were of high purity grade. Metal chlorides were Analar grade chemicals.

Preparation of the ligand.—Vanillin and ethylenediamine in stoichiometric quantities (2 : 1) were refluxed in ethanol for about two hours. The Schiff base which separated on cooling was recrystallised from ethanol and dried in vaccum over fused calcium chloride.

Preparation of the complexes.—Vanillin, ethyene diamine and the corresponding metal chlorides in appropriate molar ratio (2 : 1 : 1) were refluxed for two to three hours with ethanol. The precipitated complexes were filtered, washed with dry ethanol and dried in vacuum over fused calcium chloride.

Analysis.—The metal, nitrogen and chloride were estimated by standard procedures[3]. The results of analysis are shown in Table I.

TABLE I

Analysis of the complexes

Complex	% Metal	% Cl	% N
$ZnCl_2 \cdot L$	14·28 (14·08)	15·30 (15·27)	6·21 (6·03)
$CdCl_2 \cdot L$	21·82 (21·98)	13·95 (13·87)	5·52 (5·47)
$HgCl_2 \cdot L$	33·51 (33·39)	11·73 (11·84)	4·81 (4·67)

* The calculated values are shown in parenthesis.

Physical measurements.—The infrared spectra of the ligand and the complexes were recorded on a Carl–Zeiss spectrophotometer UR–10 (4000–400 cm^{-1}) in Nujol-mull. The ELICO conductivity bridge (cell constant 0·829) was used for conductivity measurements.

Results and Discussion

The complexes are yellow in colour soluble in DMF and DMSO and have 1 : 1 stoichiometry. Conductivity measurement indicates that the complexes are non-electrolytes.

Infrared spectra.—The medium intensity broad ligand band at 3250 cm^{-1} assignable to the intramolecular hydrogen bonded OH, existing between OH and OCH$_3$ group, is retained in the complexes also. This non-shifting of the OH band indicates the non-involvement of OH in the complex formation. Another support for this conclusion is sought in the stretching frequency due to phenolic CO. The strong band at 1290 cm^{-1} due to phenolic ν CO vibration is retained in the complexes without an alteration. Involvement of OH group in the complex formation should have shifted the ν CO to the higher frequency[4]. The strong ligand band at 1629 cm^{-1} due to ν C=N[4] appears at 1650 cm^{-1}, 1635 cm^{-1} and 1608 cm^{-1} respectively in Zn (II), Cd (II) and Hg (II) complexes. This shift may be due to the coordination through the nitrogen atom of the azomethine group. The asymmetric ν COC vibration of the ligand[5] at 1300 cm^{-1} remains inert to the complex formation, indicating non-involvement of OCH$_3$ group of the ligand in the coordination to the metal.

The involvement of both OH and OCH$_3$ groups in the bond formation would have impelled on the ligand quadridentate behavior of the type observed in the *bis*-(salicylidene) benzidene but our spectral observations show only the involvement of C=N groups in the coordinate bond formation, hence the bidentate behavior. In the 500 cm^{-1} to 400 cm^{-1} region, the medium to strong intensity bands, which are not present in the ligand, are assigned to the ν (M–N) vibrations in view of the previous assignments[6].

All these spectral observations, taken together with the analytical and conductance data, suggest that in all these complexes, the metal ions exhibit coordination number four and ligand shows bidentate behavior, coordinating through only C=N groups.

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---

1. Pujar, M. A., Hiremath, A. C. and Prabhakar, B. K., *J. Indian Chem. Soc.*, 1974, 51 (4), 494
2. Pujar, M. A., Prabhakar, B. K. and Hiremath, A. C., *Curr. Sci.*, 1976, 45 (6), 215.
3. Vogel, A. I., *A Text Book of Inorganic Analysis*, Longmans Green and Co. Ltd., London, 1962.
4. Kovacic, J. E., *Spectrochim. Acta*, 1967, 23 A, 183.
5. Rao, C. N. R., *Chemical Applications of Infrared Spectroscopy*, Academic Press, London, 1958.
6. (*a*) Prabhakaran, C. P. and Patel, C. C., *J. Inorg. Nucl. Chem.*, 1969, 31, 3316; Sharma, B. C. and Patel, C. C., *Indian J. Chem.*, 1970, 8, 747.
   (*b*) Biradar, N. S. and Kulkarni, V. H., *J. Inorg. Nucl. Chem.*, 1971, 33, 3847.

EFFECT OF *RHIZOBIUM*, *AZOTOBACTER* AND *BEIJERINCKIA* INOCULATION ON *CICER ARIETINUM* VAR. TYPE-1

Formation of nodules on the legume and the degree of nodulation are dependent on the strain of *Rhizobium* species. Therefore, a field investigation was undertaken to assess the efficiency of two strains of *Rhizobium* sps. of *Cicer arientinum*, *Azotobacter chroococcum* and *Beijerinckia indica* strains were also included in the study.

Material and Methods

A field experiment (randomized block design) with 9 treatments (Table I) replicated four times was carried out in the alluvial soil of Agricultural Farm of Banaras Hindu University. The soil pH

Table I

Showing average weight of nodules, grain yield and seed proetin of Bengal gram from a field experiment

Treatment	Wt. of dried nodules (mg/plant)	Grain yield (q/ha)	Seed crude protein (%)
Control	77	16·35	21·00
Rh. sps. H$_{45}$	146*	20·19	21·70
Rh. sps. H$_{44}$	78	17·52	20·72
Beij. indica J$_3$	103	15·98	21·35
A. chroococcum B$_4$	77	17·33	21·17
Rh. sps. H$_{44}$ + *Beij. indica* J$_3$	84	15·18	20·65
Rh. sps. H$_{44}$ + *A. chroococcum* B$_4$	203*	23·74*	21·87
Rh. sps. H$_{45}$ + *Beij. indica* J$_3$	106	20·89	21·17
Rh. sps. H$_{45}$ + *A. chroococcum* B$_4$	80	15·12	21·17
C D	66	5·19	NS

and organic matter content were 7·5 and 0·36% respectively. The plot size was 5 × 2 sq. m. The field was basal dressed with urea and superphosphate @ 20 kg N and 21·8 kg –P/ha. respectively.

Azotobacter chroococcum B_4 was isolated from B.H.U. farm soil. Strains H_{44} and H_{45} of gram *Rhizobium* and J_3 of *Beijerinckia indica* were obtained from J.N.A.U. culture collection, Jabalpur. *Rhizobium* was grown on yeast extract mannitol agar medium slants. *Azotobacter* and *Beijerinckia* were grown on Burk's medium. After an incubation period of 10 days the bacterial growth from a slant was dislodged into 100 ml sterile water for single culture treatment, but for mixed culture treatment, bacterial growth of *Rhizobium* and *Azotobacter*, or *Rhizobium* and *Beijerinckia* from two different slants was transferred to the same amount of liquid.

For seed treatment, 2 ml culture suspension and 5 ml sticker solution (2·5% sugar + 1% gum acacia) were added to 40 g seeds in a beaker and thoroughly shaken. Bacterial treatment was done just before sowing and seeds of gram (*Cicer arietinum*) var. type-1 were sown @ 40 kg/ha. After 73 days of sowing, the plants were uprooted for nodule study. At harvest the grain yield was recorded and subsequently crude protein of seeds was determined.

Results and Discussion

Data on grain yield, nodule weight, and per cent crude protein of seeds are given in Table I. Gram *Rhizobium* H_{44} caused significant increase in the nodular mass, and an increase of 23% in the grain yield which was statistically insignificant. But the same strain along with *A. chroococcum* B_4 increased the nodule weight as well as grain yield significantly over the uninoculated control. The grain yield was increased by 45%. The protein content of the seeds was not affected by bacterial inoculation.

A. chroococcum B_4 being an isolate from the same locality might have established itself when used as seed inoculant in comparison to the *Beij. indica* J_3 which was an isolate from medium black soil of Jabalpur (Sanoria, unpublished work). The beneficial effect of *Azotobacter* to *Rhizobium* and the plant might be attributed to the synthesis of growth promoting substances (Jones and Greaves, 1943; Gebgardt and Kovalchuk, 1958). Moreover, growth substance like β-indole acetic acid, possibly concerned in the nodulation process (Alexander, 1961) has been reported to be synthesized by *Azotobacter* (Smaly, 1954; Vancura and Macura, 1960). No information is available regarding the production of growth substances by *Beijerinckia*. Possibly *Beijerinckia* which produces abundant slime, excretes little of the growth substances. Both the strains H_{44} and H_{45} were reported to be effective under the medium black soil of Jabalpur (Sanoria and Dube, 1972) but in the Indo-gangetic alluvial soil of Varanasi only H_{44} is found to be effective. Local strain B_4 is found to exert synergistic effect on H_{44}.

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1. Alexander, M., *Introduction to Soil Microbiology*, John Wiley and Sons, Inc., New York and London, 1961, p. 332.
2. Gebgardt, A. G. and Kovalchuk, S. I., *Mikrobiol.*, 1958, 27, 331.
3. Jones, L. W. and Greaves, J. E., *Soil. Sci.*, 1943, 55, 393.
4. Sanoria, C. L. and Dube, J. N., *Rhizobium News Letter*, 1972, No. 1.
5. Smaly, V. T., *Mikrobiol. Zh.*, 1954, 9, 17.
6. Vancura, V. and Macura, J., *Folia Microbiol.*, 1960, 5, 293.

RHYTHMIC VARIATIONS IN THE PHOSPHORYLASE ACTIVITY IN THE SCORPION *HETEROMETRUS FULVIPES* (KOCH)

DIURNAL rhythms in various activities like locomotion, poison secretion[1], neurosecretion[2], rate of heart beat, cholinesterase activity in the heart muscle[3], spontaneous electrical activity in the ventral nerve cord and segmental nerves[4], have been reported to occur in the scorpion *Heterometrus fulvipes*. Similar rhythms have also been shown to occur in the levels of metabolites like blood glucose and hepatopancreatic glycogen in the same species[5]. Carbohydrate distribution and synthesis were studied in different tissues of scorpion[6]. The present report concerns the study of phosphorylase activity in the pedipalpal muscle and hepatopancreas of the scorpion *H. fulvipes* as a function of time of the day. The pattern of activity of this enzyme which plays a vital role in glycogen breakdown should reveal the pattern of utilization of carbohydrate energy sources for various activities during the course of a 24 h period.

The details of collection, maintenance of scorpions and sampling of tissues were described earlier[5,6]. The activities of phosphorylase '*a*' (active) and '*ab*' (total) have been estimated in the absence and presence of AMP respectively according to the method described by Cori, Illingworth and Keller (1955)[7]. A 5% homogenate was prepared in aqueous medium containing 0·037 M ethylene diamine tetra acetic acid (EDTA), pH 6·5 and 0·1 M sodium fluoride, pH 6·5, as recommended by Guillory and Mommaerts[8]. The inorganic phosphate was estimated by the method of Taussaky and Shorr[9] and protein concentration was estimated by the method of Lowry *et al.*[10].

The results presented in Fig. 1 show that the total phosphorylase ('*ab*') ranges from 25·73 to 31·0 μ moles of Pi/mg protein/h in the pedipalpal muscle and 23·1 to 32·7 μ moles of Pi/mg protein/h in hepatopancreas. The range is essentially similar even though statistical treatment shows that at 04·00 h and 08·00 h the total phosphorylase activity in muscle is significantly slightly higher ($P < 0{\cdot}05$) than that of hepatopancreas. In both the tissues the maximal activity was recorded at 20·00 h and the minimal activity was recorded at 08·00 h. But the pattern of rise and fall in total phosphorylase activity is different in the two tissues studied. In the muscle the rise is less pronounced. Only the peak period value at 20·00 h is significantly higher than the value at 08·00 h ($P < 0{\cdot}002$). The decline also is gradual, the midnight value being not significantly different from 20·00 h value ($P > 0{\cdot}8$) which the 04·00 h is significantly lower than 20·00 h value ($P < 0{\cdot}05$). In the hepatopancreas the rise and fall in enzyme activity are more steeper, the value at each period being significantly different from the value at the previous interval.

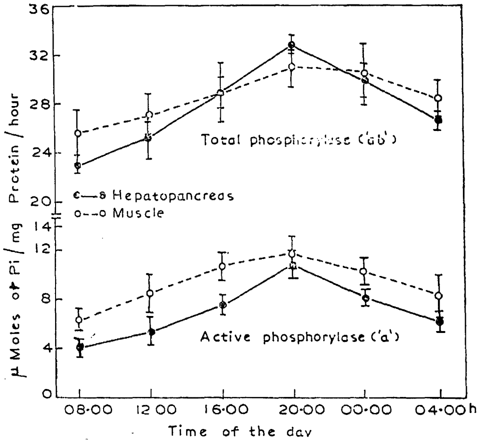

FIG. 1. Phosphorylase activity in the scorpion *Heterometrus fulvipes* as a function of time of the day. Each point represents the mean of six estimations ± S.D.

The general pattern of activity of phosphorylase '*a*' is very similar to that of total phosphorylase, the activity being maximal at 20·00 h and minimal at 08·00 h in both the tissues. It ranges from 6·06 to 11·53 μ moles Pi/mg protein/h in the muscle and from 4·05 to 10·67 μ moles Pi/mg protein/h in the hepatopancreas. Muscle active phosphorylase is consistently higher than that of hepatopancreas. In the muscle even though the peak enzyme activity is found at 20·00 h, the enzyme activity keeps continuously high between 16·00 h and 0·00 h. In hepatopancreas the rise and fall are more sharper.

The higher phosphorylase '*a*' activity in the muscle perhaps reflects glucose utilization for muscle contraction. Hepatopancreas is known to be the main organ of storage of nutrients. Synthesis and breakdown of glycogen in hepatopancreas are related to general metabolic needs of the animal. Both active and total phosphorylase activities were maximal at 20·00 h. It was reported earlier that blood glucose value was maximal at 20·00 h while the hepatopancreas glycogen content was minimal indicating mobilization of carbohydrate from hepatopancreas to blood[5]. The maximal phosphorylase activity reported in the present investigation adequately explains the earlier results. The minimal phosphorylase activity at 08·00 h is correlated to the maximal glycogen value in hepatopancreas and minimal blood sugar level. The pattern of variation in phosphorylase activity is of interest in the context of rhythmic patterns seen in other activities and the behaviour of the scorpion. The scorpion is a nocturnal creature having high locomotor activity during the night times[1]. The metabolic rate also is high between 16·00 h and midnight (0·00 h) with a peak around 20·00 h[1]. We have observed that the scorpions remain apparently inactive lurking under the stones during the most of the day time. They exhibit active feeding behaviour during the night time. Rate of heart beat and cholinesterase activity of the heart muscle are also higher during the dark hours with a peak activity at about 20·00 h[3]. The spontaneous electrical activity in the ventral nerve cord is reported to be maximum at 16·00 h while the electrical activity in the segmental nerves is maximal at 20·00 h[4]. The high phosphorylase activity in the scorpion during the night period at and around 20·00 h should be providing the necessary glucose from glycogenolysis, required for energetic needs of various physiological activities. This is particularly true of hepatopancreas phosphorylase, since the glycogen in this tissue is a more labile store of energy. Muscle phosphorylase, on the other hand, augments glycogenolysis yielding glucose which would be mostly utilized as energy source for contraction process. After 20·00 h phosphorylase activity decreases as there would be enough glucose in the blood due to post-prandial absorption. The excess glucose is perhaps transported to hepatopancreas for glycogen synthesis. This is evident from the observation that the glycogen level in hepatopancreas gradually increases after 20·00 h reaching a maximal value at 08·00 h in the morning[5].

It is known in mammals that epinoephrine induces greater production of cyclic AMP in muscle resulting in increase of active form of phosphorylase which in turn augments glycogenolysis. It is of interest that a hyperglycemic principle has been identified in the extracts of scorpion cephalothrocic nerve mass[11]. Circadian rhythms in the neurosecretory activity in the scorpion has also been reported[2]. It is probable that the active principle from scorpion neurosecretory system with epinephrine-like action is responsible for activating the phosphorylase system.

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1. Gopalakrishna Reddy, T., *Doctoral Dissertation* submitted to Sri Venkateswara University, Tirupati, 1966.
2. Habimullah, Md., *Doctoral Dissertation* submitted to Sri. Venkateswara University, Tirupati, 1966.
3. Devarajula Naidu, V., *Experientia*, 1969, 25, 1274.
4. Venkatachari, S. A. T., *Ind. J. Exp. Biol.*, 1971, 9, 338.
5. Chengal Raju, D., Bashamohideen, M. D. and Narasimham, C., *Experientia*, 1973, 29, 964.
6. Ramamurthi, R., Satyanarayana, K., Selvarajan, V. R. and Swami, K. S., *Ind. J. Exp. Biol.*, 1975, 13 (5), 446.
7. Cori, G. T., Illingworth, B. and Keller, P. G., In: *Methods in Enzymology*, Eds., Colowick, S. P. and Kaplan, N. O., Academic Press, New York, Vol. I, p. 200.
8. Guillorv, R. J. and Momeaerts, W. F., H.M., *Biochem. Biophys. Acta*, 1962, 65, 316.
9. Taussaky, H. M. and Shorr, E., *J. Biol. Chem.*, 1953, 202, 675.
10. Lowry, O. H., Rosbrough, N. J., Farr, A. L. and Randall, R. J., *Ibid.*, 1951, 193, 265.
11. Raghavaiah, K. and Ramamurthi, R., *1st All India Symposium on Comparative Animal Physiology*, Aurangabad, November 1975. Abstract Volume, p. 46.

RETRACTOCEPHALUS—A NEW GENUS OF CEPHALINE GREGARINES (PROTOZOA: SPOROZOA) FROM INSECTS

Kudo[1], recognised 12 genera of cephaline gregarines (Protozoa: Sporozoa) under the family Gregarinidae Labbé, 1899. In our studies on this group of parasites from insects of this locality, we have found that the cephaline gregarines inhabiting the various parts of the mid gut of the beetles, belonging to the family Chrysomelidae possess sporonts in syzygy, simple globular retractile epimerite, cysts without ducts and barrel-shaped spores extruded in chains—characters absent in any known genus of the family Gregarinidae. A new genus, *Retractocephalus* is, therefore, proposed here to accommodate these gregarines. The genus is characterised by the following diagnostic features:

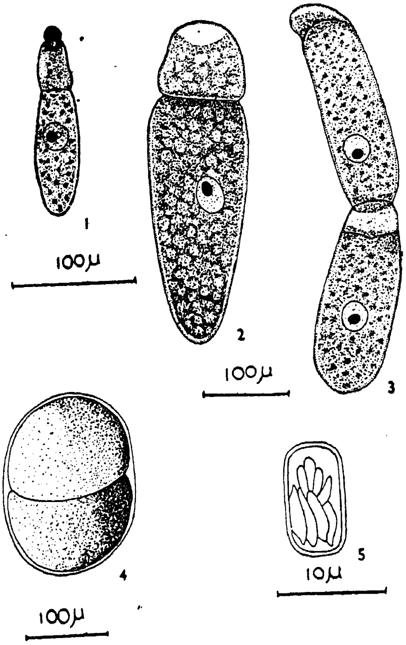
(1) initial development of the parasite is intracellular;
(2) sporonts are in syzygy and the association is caudo-frontal in nature;
(3) the epimerite is a simple symmetrical globular structure and retractile into the protomerite;
(4) dehiscence of the cyst is by simple rupture;
(5) the spores are barrel-shaped and extruded in chains.

The gregarine, *Retractocephalus raphidopalpii* obtained from the mid gut of the beetle, *Raphidopalpa* (= *Aulacophora*) *foveicollis* (Lucus) is designated as the type species of the genus. A brief description of the organism is given here. Details of its morphology and life history will be dealt with separately.

The parasite undergoes its initial development within the epithelial cells of mid gut of the host. With the development of the epimerite, it leaves the infected cell and remains attached with it by the epimerite for some time. Later, it frees itself from there, begins to live inside the gut lumen. A fully grown trophozoite (Fig. 1) has an elongated body. Its epimerite is a simple hyaline globular structure and occasionally retracts completely into the protomerite. The protomerite is elongated, cylindrical and has a depression to receive the epimerite. The deutomerite is the largest segment of the body and is separated from the protomerite by a thick straight septum. The nucleus is spherical or slightly oval enclosing one or two karyosomes.

The sporonts (Fig. 2) are larger in size than the trophozoites and an epimerite is wanting at this stage. The sporonts associate caudo-frontally (Fig. 3) when the posterior tip of the primite fits firmly into a concavity of the protomerite of the satellite.

A freshly collected gametocyst from the hind gut of the host is oval, opaque white and encloses two equal gametocytes (Fig. 4). The cyst measures 250·0 × 210·0 μ to 310·0 × 250·0 μ in dimensions. At 48 hours of development the cyst dehisces by normal rupture. The spores are barrel-shaped and are extruded in chains. These measure 7·0 × 4·0 μ and have a double wall. Formation of eight sporozoites is completed at 70 hours of development (Fig. 5).



FIGS. 1–5. Camera lucida drawings of *Retractocephalus repahidopalpii* n. gen., n. sp. Fig. 1. A trophozoite. Fig. 2. A sporont. Fig. 3. Sporonts in syzygy. Fig. 4. A freshly collected gametocyst enclosing two gametocytes. Fig. 5. A fully developed spore with 8 sporozoites inside.

Measurements (in microns) :

(*i*) Trophozoites—LE 7·5 to 15·0 (average 10·0) ; LP 15·0 to 32·5 (average 26·5) ; LD 35·0 to 92·5 (average 83·0) ; TL 57·5 to 135·0 (average 116·5) ; WP 17·5 to 27·5 (average 22·0) ; WD 20·0 to 37·5 (average 28·5) ; nucleus 12·5 to 22·5 (average 16·0).

(*ii*) Sporonts—LP 22·5 to 85·0 (average 36·8) ; LD 65·0 to 290·0 (average 147·4) ; TL 87·5 to 360·0 (average 183·6) ; WP 20·0 to 100·0 (average 34·2) ; WD 25·0 to 120·0 (average 44·2) ; nucleus 10·0 to 45·0 (average 22·7).

LP : TL = 1 : 4·9 ; WP : WD = 1 : 1·2.

The generic name *Retractocephalus* has been given to emphasize the retractile nature of the epimerite, and the genus denotes a neuter gender. The specific trivial name has been derived from the generic name of the host of the type species. The type materials have been deposited at the Zoology Department, Kalyani University.

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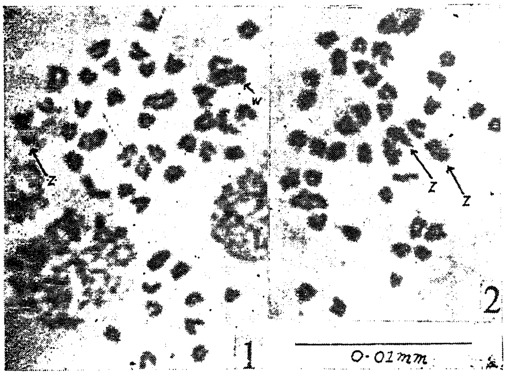
1. Kudo, R. R., *Protozoology*, 5th ed., Charles C. Thomas, Illinois.

CYTOLOGICAL FEMALE HETEROGAMETY IN JET-BLACK MOLLY, *MOLLIENESIA SPHENOPS*

EARLY workers on fish material could not find any morphologically distinguishable sex chromosomes mainly because of two reasons. Firstly, their techniques revealed little details of the chromosome morphology and secondly, their studies were mostly limited to the male germinal chromosomes or to the somatic chromosomes of immature, sexually undifferentiated specimens (*cf.* Chiarelli and Capanna)[1]. Recently, however, many workers have begun exploiting the colchicinized somatic tissues from both the sexes, and with this, there have appeared quite a few reports on the occurrence of heteromorphic sex chromosome pairs in a number of teleost fishes[2-7]. Even multiple sex chromosomes, along with the confirmation of the sex multivalent formation during meiosis, have been detected in three species[8-10]. Therefore, it is not unlikely that fishes, on being cytologically exploited on a more elaborate scale, would reveal a more widespread cytologically expressed heterogamety than previously believed.

A case of cytologically expressed heterogamety has been observed by us in the jet-black molly, *Mollienesia sphenops* (Cuv. and Val.), a cyprinodontid, kept in a well aerated aquarium. For study, each specimen received a 0·01 ml intraperitoneal injection of 0·1% colchicine. After two hours, the gill arches were removed and pretreated with 1·0% sodium citrate solution for 25 min. and fixed in 1 : 3 acetic alcohol for 30 minutes. The gill arches were stained in Gomori's haematoxylin after Melander and Wingstrand[11]. After differentiation in 45% acetic acid, the gill epithelium was taken in small bits and squashed. Permanent slides were prepared by freezing the slides in a refrigerator. Similarly, squash preparations were also made from the spleen and kidneys.

After the scrutiny of a number of metaphases from the three somatic tissues, it is found that the karyotype of this fish in both the sexes comprises of 46 chromosomes. However, the female complement has 45 acrocentric elements and one large metacentric (Fig. 1) whereas the male complement is marked by all acrocentric chromosomes (Fig. 2). The occurrence of the large metacentric is confirmed in a number of female specimens and in all the three tissues. This, therefore, eliminates the possibility of this being a case of usual karyotypic polymorphism, so commonly found in teleosts[12-15]. In the female metaphases, in addition to the lone big metacentric, there is a lone acrocentric (the smallest). We, therefore, propose these two elements to comprise the WZ pair of sex elements. On the other hand, in the male complement there is a pair of small acrocentrics (the smallest), which have been labelled as ZZ sex elements. These sex elements in the female as well as in the male can be recognized even without resorting to karyotyping. Hence, the female sex in *M. sphenops* is clearly heterogametic. Due to the undeveloped gonads in our specimens (due to non-breeding season), we could not confirm the behaviour of the sex chromosomes during meiosis. There was, however, no difficulty in recognizing the sexes because of sexual dimorphism.



FIGS. 1–2. Fig. 1. Female metaphase from gill epithelium. Fig. 2. Male metaphase from kidney.

The cyprinodontids show a clear sexual dimorphism and are well reputed for the genetic analyses of their sex chromosomes (*cf.* Kallman)[16]. A well-authenticated report about the cytological confirmation of the presence of sex chromosomes is also on record by Chen and Ebeling[3]. They have found in the mosquito fish, *Gambusia affinis,* a similar cytological heterogamety as in our present material. In view of these observations, a more thorough cytological survey of the family Cyprinodontidae as a whole is well warranted.

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1. Chiarelli, A. B. and Capanna, E., *Cytotaxonomy and Vertebrate Evolution,* Acad. Press, London, 1973.
2. Chen, T. R. and Ebeling, A. W., *Chromosoma,* 1966, 18, 88.
3. — and —, *Copeia,* 1968, 1, 70.
4. —, *Postilla,* 1969, 130, 1.
5. Ebeling, A. W. and Chen, T. R., *Trans. Amer. Fish. Soc.,* 1970, 99, 131.
6. — and Setzer, P. Y., *Copeia,* 1971, 3, 560.
7. Rishi, K. K., *Genen Phaenen,* 1973, 16, 101.
8. Uyeno, T. and Miller, R. R., *Nature,* 1971, 231, 452.
9. — and —, *Experientia,* 1972, 28, 223.
10. Rishi, K. K., *Ciencia E, Cultura* (Communicated).
11. Melander, Y. and Wingstrand, K. G., *Stain Technol.,* 1953, 28, 217.
12. Ohno, S., Stenius, C., Faisst, E. and Zenzes, M. T., *Cytogenetics,* 1965, 4, 117.
13. — and Atkin, N. B., *Chromosoma,* 1966, 18, 455.
14. Becak, W., Becak, M. L. and Ohno, S., *Cytogenetics,* 1966, 5, 313.
15. Roberts, F. L., *Can. J. Genet. Cytol.,* 1968, 10, 865.
16. Kallman, K. D., *Zoologica.,* N.Y., 1965, 50, 151.

RECORD OF *PTEROLOPHIA ANNULATA* CHEVR. AND *DIBOMA PROCERA* PASC. (LAMIINAE; CERAMBYCIDAE: COLEOPTERA) AS NEW PESTS OF PEPPER (*PIPER NIGRUM* L.) FROM INDIA

IN black pepper (*Piper nigrum* L.) about 20 species of insects have been reported as pests so far. Of these 'Pollu' beetle (*Longitarsus nigripennis* Mots.)[1] is the most important one. During a routine survey of the pepper growing tracts of the northern parts of Kerala in the summer months of 1975, some pepper vines in the Alakode area were found severely affected by stem borer pests causing the death of the affected vines. Similar infestations were noticed in pepper plantations in other localities of Cannanore and Calicut Districts. The incidence of the pests was as high as 50% on vines in some gardens. Preliminary observations are reported in this note.

Grubs of the longicorn beetles bore into the stem causing damage to the vines. They tunnel into and feed on the central core of the stem. The tunnels are tightly packed with frass as the grub progresses forward. A fully developed grub is 12–18 mm long and has a dull yellow colour. It has a swollen anterior end and a tapering posterior end. Before pupation, the grub makes an exist hole on the stem and then pupates inside a pupal chamber and the adult beetle makes its way out easily. The grubs were often found in the older and dead vines. They were also noticed tunnelling into the living vines at the collar region or slightly below. However, the grubs have preference to the dead and dried tissues.

The beetles reared from the affected pepper vines have been identified by the Commonwealth Institute of Entomology, London, as *Pterolophia annulata* Chevr. and *Diboma procera* Pasc. (Lamiinae ; Cerambycidae : Coleoptera).

From the symptoms of the affected vines in Travancore and Coorg areas, Krishna Menon[4] reported that a borer is involved in the damage though he did not observe the presence of grubs and adults in the affected vines. Blacklock[3] recorded grubs of the beetle pest *Pelargoderus bipunctatus* Dalm. infesting the stems of unhealthy pepper vines in Sarawak.

This is the first record of the above insects as pests of pepper. Another recorded host of *D. procera* Pasc. is *Acacia gageana*[2], *P. annulata* Chevr. has not been recorded from India so far. However, allied species like *P. tuberculata* and *P. maculata* have been recorded from *Tectona grandis* and *Michelia champaka* respectively[2].

We express our sincere thanks to Drs. Y. R. Sarma and K. K. N. Nambiar, for drawing our attention to these pests. We are also grateful to Dr. A. H. Parker, Ag. Director, Commonwealth Institute of Entomoloby, London, for identification of pests.

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1. Ayyar, T. V. R., *Hank Book of Economic Entomology for South India*, Government Press, Madras, 1940. p. 442.
2. Beeson, C. F. C., *The Ecology and Control of the Forest Insects of India and the Neighbouring Countries*, 1941. p. 767.
3. Blacklock, J. S., *Trop. Agr.* (*Trin.*), 1954, 31 (1), 40.
4. Krishna Menon. K., *Indian J. Agr. Sci.*, 1949, 19 (1), 89.

AN UNDESCRIBED SPECIES OF *CERCOSPORA* CAUSING LEAF SPOT OF *MITRAGYNA PARVIFOLIA* (ROXB.) KORTH.

INFECTED leaves of *Mitragyna parvifolia* (Rubiaceae) were seen having reddish brown to tan brown spots during winter. The spots were seen occurring on the upper surface of the leaf and had a tendency to coalesce with each other to form bigger spots of different shapes and sizes specially towards the margins of the leaf. The material was also examined and confirmed by Dr. Mulder of C.M.I., Kew, to whom our thanks are due.

Cercospora mitragynae sp. nov. Bhargava and Nath.

Leaf spots chiefly circular, 2–3 mm. in diameter, chocolate in colour, epiphyllous on the upper surface. Greenish brown areas are visible on the lower surface of the leaves, corresponding with the leaf spots above. Fruiting epiphyllous. Stroma subglobose composed of dark brown cells, 30 μ in diameter. Fascicles dense, fairly compact. Conidiophores pale to medium olivaceous brown, arising in fascicles of 9–10, non septate occasionally branched, sinuate towards apex, tip hyaline with small spore scar, 15–50 × 2·5–3·5 μ. Conidia olivaceous brown, obclavate or subcylindric, sometimes vermiform, slightly curved at the centre, 6–14 septate, 25–75 × 2·5–3·3 μ (Fig. 1).

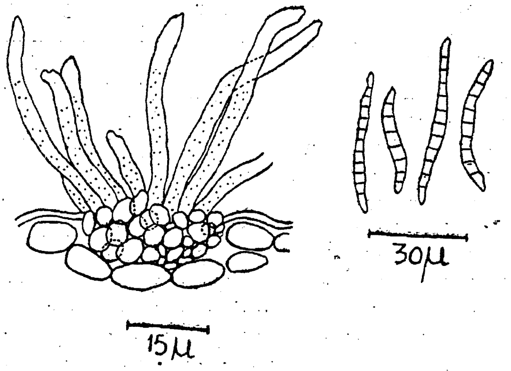

FIG. 1. *Cercospora mitragynae* sp. nov. on *Mitragyna parvifolia*, Sporodochia, Conidiophores and Conidia.

Cercospora mitragynae Bhargava *et* Nath, sp. nov.

Maculae foliorum praesertim circulares, 2–3 mm diam., badii, epiphylli, quibus congruentes infra sunt areae viridibrunneae. Fructificatio epiphylla. Stroma, cellulis 30 μ diam. atrobrunneis, subglobosum. Fasciculus densus, satis compactus. Conidiophora pallida ad medio-olivaceo-brunnea, fasciculis 9–10 orientia, non-septata, interdum ramosa, ad apicem sinuata 15–50 × 2·5–3·5 μ; apex hyalinus, cicatrice sporae parva. Conidia olivaceo-brunnea, obclavata vel subcylindrica, interdum vermiforma, leviter ad centrum curvata, 6–14 septata, 25–75 × 2·5–3·3 μ.

Hab.—On living leaves of *Mitragyna parvifolia* (Roxb.) Korth., Kushmi and Madhaulia forest, Gorakhpur, India. November, 1974 leg. V. Nath. Type specimen of infected leaves deposited at C.M.I., Kew (No. IMI, 191340) and the isotype at the Botany Department, Gorakhpur University, Gorakhpur, India.

The species differs from *C. adinae* in having epiphyllous leaf spots, in not having geniculate conidiophores, the apex and the shape of conidia. This also differs from *C. ixorae* in the appearance of leaf spots, in not having hypophyllous conidiophores with more or less a bulbous tuberculate base, and cylindrical and subhyaline conidia.

The authors are thankful to Dr. K. M. Matthew, S.J., St. Joseph's College, Tiruchirapalli, for rendering the text in latin.

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OBSERVATIONS ON THE STOMATAL MORPHOLOGY OF LEAF GALLS OF *RIVEA HYPOCRATERIFORMIS* CHOISY FORMED BY *ASPHONDYLIA RIVEAE* MANI

STOMATAL abnormalities in relation to galling as well as various morphological anomalies exist in many galls induced by fungi[1,2] and by insects[3-6]. Though detailed anatomical studies are available[7] on the fleshy epiphyllous galls on the leaves of *Rivea hypocrateriformis*, information on the morphology of the stomata appear wanting. Hence, an attempt is made here to present the stomatal anomalies caused under the influence of the gall maker.

Normal and galled leaves of *Rivea* were macerated in Jeffrey's fluid to obtain the epidermal peels, which were stained with iron alum-haematoxylin and mounted in glycerin jelly. Normal leaves bear paracytic stomata (Fig. 1 A) along the lower surface and the stomata are 40–45 × 30–35 μ in dimension. The upper ledge is uniformly thick and the stomatal index is 12·1. Significant anomalies (Fig. 1 B–K) were evident in the stomata which occur only on the lower sides of the galled leaves. The distribution of the stomata appears quite irregular, being concentrated along certain areas of the epidermis while sparse or even completely absent in others with the stomatal index declining to 6·6. Both smaller (Fig. 1 B) and

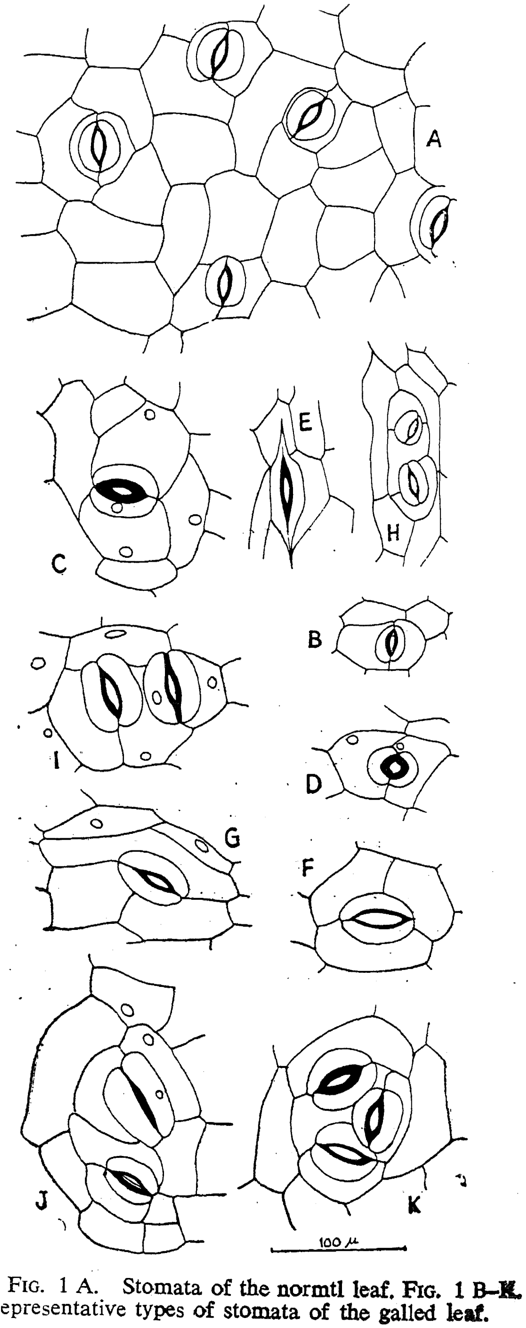

FIG. 1 A. Stomata of the normtl leaf. FIG. 1 B–K. Representative types of stomata of the galled leaf.

larger stomata (Fig. 1 C) were evident measuring 23–25 × 18–20 μ and 50–55 × 47–50 μ respectively. Stomata with highly elongate guard cells (Fig. 1 E) were observed towards basal region of the galls, *i.e.*, towards the midrib. The stomata appear curiously smaller along the mid region of the gall with a characteristic circular outline (Fig. 1 D). On specific stomata, the upper ledges were enormously thickened. Contrary to the normal occurrence of two subsidiary cells, three subsidiary cells were also observed (Fig. 1 F and G). Stomata occur in closely agglomerated groups of two sharing a common subsidiary cell (Fig. 1 H, I and J). In an extreme case, three stomata occurred in very close proximity, that the guard cells were almost abutting on each other (Fig. 1 K).

Observations on the anomalous stomata of *Rivea* galls indicate that they are functional, but for the change in morphology and distributional patterns. Fall in the stomatal index is significant, since there is an enormous growth resulting in the gall. Effects of wounding and hormone treatments have been known to inhibit stomatal development, and the occurrence of stomata of lower stomatal index on *Rivea* galls is in conformity to the basic principles of gall formation, which involve wounding and hormone effects through insect action.

Prof., T. N. Ananthakrishnan, Layola College, Madras, is gratefully thanked for the critical reading of the manuscript. One of us (A. R.) thanks the University of Madras, for the award of the University Teacher Fellowship.

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Madras-59, *December* 27, 1975.

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1. Akai, S., *Mem. Coll. Agriculture, Kyoto Univ.*, 1951, **58**, 1.
2. Meyer, J., *Bull. de la Soc. bot. Fr.*, 1955, **102**, 6.
3. Küster, E., *Anatomie der Gallen*, Handb. der Pflanzenanatomie, Ed. Linsbauer, K., 1930.
4. Meyer, J., *Marcellia*, 1959, **30**, 95.
5. Schmidt, E. and Meyer, J., *Ibid.*, 1966, **33**, 237.
6. Krishnamurthy, K. V. and Raman, A., *Ibid.*, 1972, **37** (5), 21.
7. Mani, M. S., "Cecidotheca Indica," *Agra Univ. J. Res.* (*Sci.*), 1959, **8**, 91.

OCCURRENCE OF A NEW RACE OF *HELMINTHOSPORIUM MAYDIS* ON MAIZE

During the month of September, 1975 a very severe peculiar leaf blight of maize (Fig. 1) was observed on two hybrids (EH 2310 and EH 2420) at Punjab Agricultural University, Ludhiana. The diseased specimens were examined microscopically and were found to be associated with *Helminthosporium maydis*. Brown blotches were observed on leaf sheaths and husk covers, underneath which the kernels showed dull black appearance because of rotting (Figs. 2 and 3). The isolation of infected grains also yielded *H. maydis*. These hybrids were under strict pathological evaluation and observed to be tolerant to the prevalent race 0, of *H. maydis*[1]. Moreover, the symptoms were quite different from that of leaf blight caused by race 0, which causes small diamond-shaped lesions on leaf lemina[2]. These facts warranted further study of this disease.

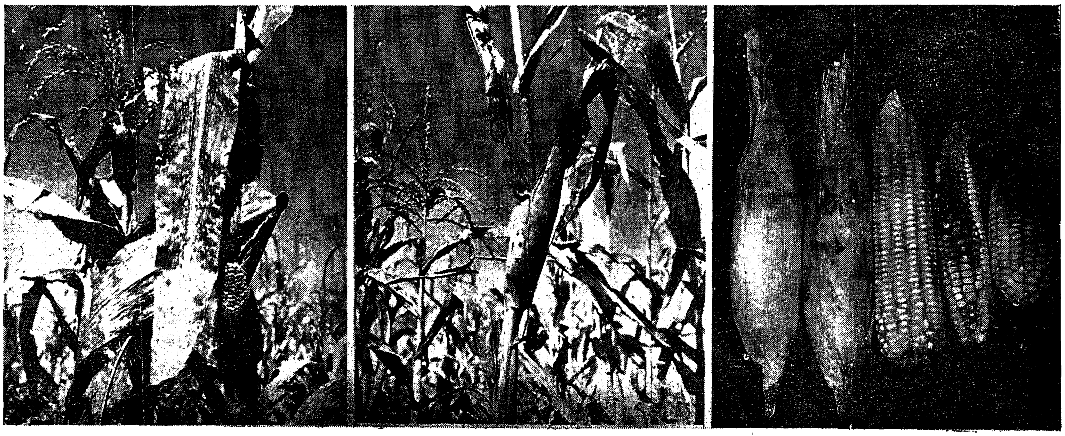

Fig. 1 Fig. 2 *Fig. 3

Figs. 1–3. Disease symptoms on leaf lamina, leaf sheath, ear husks and kernels (*Fig. 3—Middle one is healthy ear).

The seedlings of EH 2310 and EH 2420 were inoculated with spore suspension. Within 6–7 days, the fungus induced symptoms of blight quickly. Cobs at milk stage were inoculated by keeping the culture bits underneath the husk covers. The blotches on husk cover and kernel rotting were observed after 7 days of inoculation. The infection type produced on leaves, husks and kernels was same as observed in field and resembled to race T[2], which caused havoc in U.S.A. in 1970[3]. This race was reported to be specifically adapted to 'Texas male sterile' cytoplasm[2]. However, varieties with normal cytoplasm were also later on reported to be susceptible[4], as is the case with present hybrids. This is first record of such a virulent race in India on maize which is being compared with T race and details will be reported later. It is worth mentioning that T race had been recorded in India on sorghum[5].

Under field conditions, almost 100% of plants were infected and the ear and grain development was very badly affected (Fig. 3) resulting in drastic reduction in yield. In comparison to standard hybrid (Ganga 5 = 3250 kg/ha), the grain yield of these hybrids was lower by 30 to 40%.

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1. Anonymous, *Progress Report, Coordinated Maize Improvement Scheme, Section of Plant Pathology,* I.C.A.R., New Delhi, 1974–75.
2. Hooker, A. L., Smith, D. R., Lim, S. M. and Musson, M. D., *Plant Dis. Reptr.,* 1970, 54, 1109.
3. Ullstrup, A. J., *Ann. Rev. Phytopath.,* 1972, 10, 37.
4. Craig, J., *Plant Dis. Reptr.,* 1971, 55, 627.
5. Nath, R., Lambat, A. K., Payak, M. M., Lilaramani, J. and Rani, I., *Curr. Sci.,* 1973, 42, 872.

SHORT SCIENTIFIC NOTES

Induction of Amber-grained Mutants in Bread Wheat Var. Inia-66

Inia-66, a bread wheat variety (L. Rojo-64 × Sonora-64) introduced to India from Mexico, atthough possesses very high yield potentiality could not be accepted for commercial cultivation due to the drawback of having red-grained type. With a view to rectify this defect, induction of mutants having amber-grain colour was attempted with the use of gamma radiation (30 kR and 40 kR) and combination of radiation and ethyl methane sulphonate (30 kR + 0·2% EMS). . The frequency of occurrence of amber-grained mutants in M_2 generation is shown in Table I. All the mutants bred true for amber-grain colour in subsequent generations. Except for the grain colour, the mutants were having identical morphological characteristics and equally high yielding potentiality like that of parental variety. Moreover, three amber-grained mutants have shown trace resistance to brown rust whereas the parental variety is highly susceptible to brown rust. In the evaluation trials the commercial potentiality of an amber-grained mutant could be established.

Table I

Frequency of amber-grained mutants in M_2 generation in Inia–66

Treatment	No. of M_1 plants studied	Nos. of M_2 plants studied	Nos. of mutants isolated	Frequency of mutants per 100 M_2 plants
Control	150	4950	..	..
30 kR	120	4500	3	0·07
40 kR	30	1200	..	0·00
30 kR + 0·2% EMS	150	4050	3	0·07

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Colorimetric Estimation of Iron with Haematoxylin

Haematoxylin [3, 7, 8, 5′, 6′,-Pentoxy (indeno 2′, 1-3, 4-Chomene)-dihyride-(3, 4)] has been used for the colorimetric determination of aluminium, boron, niobium, tin, lanthanides[1] and is a metallochromic indicator for aluminium, bismuth, copper thorium and zirconium[3]. It's reaction with iron (III) has also been studied[4].

Haematoxylin forms a bluish complex with iron (III) in the pH range 5·5–6·0 with acetate buffer. This reaction has been found to be most satisfactory for the estimation of trace amounts of iron in marble, magnesite and similar materials. The standard graph plotted between known concentrations of iron against optical density at wavelength 530 nm gave a straight line.

Experimental

Sample of marble and magnasite were decomposed and brought into solution. Aluminium was separated by treating the solution with excess of NaOH. Precipitate of Fe $(OH)_3$ was dissolved in HCl. To this solution, acetate buffer and Haematoxylin solution were added to develop the blue colour. The optical density was measured at 530 nm. using an sp. 600 Spectrophotometer. The results obtained by this method compare favourably with those obtained by conventional methods[2,5].

Beer's law is found to be obeyed up to 40 μg/ml. At higher concentrations, a suspension is formed and it is advisable to work at concentrations below 40 μg/ml. The reagent solution is stable for about five days. Aluminium and fluoride interfere seriously while nickel, cobalt, mercury and thorium interfere to a small extent. Ca and Mg do not intefere at this pH.

The authors' thanks are due to the Principal, P.P.N. College, Kanpur and the Director, Geology and Mining, U.P., Lucknow, for providing laboratory facilities.

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---

1. Malet, M., *Absorpanei Anorganicke Photometric,* Academia Praha, 1973.
2. West, T. S., *Complexometry with EDTA and Related Reagents,* 3rd Ed., 1969, p. 54.
3. Bishop, E. (ed.), *Indicators,* Peigamori, Oxford, 1972.
4. Chandra, S., Sahgal, R. K. and Prakash, D., *L.A.B.P.,* 1975, 26, 347.
5. Sandell, E. B., *Colorimetric Determination of Traces of Metals,* Inter-Science Publishers, Third Ed., 1958, p. 242.

Chromosomal Status of Vellai-columban (*Mangifera indica* L.)

The chromosome number in *Mangifera indica* L. has been observed to be $2n = 40$[1]. An exception to this rule was reported by Roy and Visweswariya[2] in the case of 'Vellai Columban', a polyembryonic variety of mango which was found to be a tetraploid (Quoted by Singh)[3]. This variety appeared to be of practical significance in evolving triploid and aneuploid types in mango. However, before utilizing this material in the breeding programme it was thought desirable to confirm its chromosome number. The observations made are presented in this note.

The material for cytological studies was obtained from the same source as that of Roy and Visweswariya[2]. The flower buds from just emerging panicles were fixed between 10 to 11 a.m. in modified carnoy (6 alcohol : 3 acetic acid : 1 chloroform) to which a few drops of saturated Ferric chloride were added. The anthers were squashed in 2% acetocarmine.

The meiotic chromosomes were counted from diakinesis and early metaphase. Examination of a large number of preparations invariably revealed only 20 bivalents. There were no quadrivalent associations or abnormal anaphases. The acetocarmine stainability tests also showed a very high degree of pollen fertility (95%). These observations reveal that the variety Vellai Columban is a diploid ($2n = 40$) and not a tetraploid, as has been reported in the literature so far.

The authors are thankful to Dr. R. N. Singh, Head of the Division of Horticulture and Fruit Technology, I.A.R.I., New Delhi, for providing the necessary facilities.

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---

1. Mukherjee, S. K., *Proc. Nat. Inst. Sci., India,* 1950, 16, 287.
2. Roy, B. and Visweswariya, S. S., *Maharashtra Assoc. for Cultivation of Sci., Poona,* 1951.
3. Singh, L. B., *The Mango,* Leonard Hill, London, 1960.

Conchyliurus bhimilensis N. Sp. (Copepoda) from a Bivalve Mollusc *Meretrix casta* (Chem) off Bhimunipatnam

Information on Copepods parasitic on marine organisms other than fishes is scanty. It was therefore of some interest to observe a copepod of the genus *Conchiliurus* inhabiting the mantle cavity of a bivalve mollusc *Meretrix casta.* Of over 100 specimens examined, 10 harboured the copepod. In a few, more than one parasite was noticed. Of the 14 parasites collected 10 were females, and of these, two had ovisacs.

Conchyliurus has been described earlier from *Meretrix.* Reddiah[1,2] found two distinct species, *C. bombasticus* and *C. fragilis* from *Meretrix meretrix.* The present specimen of *Conchyliurus* differs from both *C. bombasticus* and *C. fragilis* in the following respects (*see* Table I). Since it is felt the

TABLE I

Character	*C. bombasticus*	*C. fragilis*	*C. bhimilensis*
Colour of the live specimen	Colourless with brick-red eyespot	Colourless with brick-red eyespot	Pink, with dark red eyespot spermatophore orange brown
Size of spermatophore	0·25 mm	Did'nt mention	0·176 mm
Length of Ovisac	0·8 × 0·2 mm	0·65 × 0·2 mm	0·704 × 0·160 mm
No. of eggs	60–70	35 to 40	69–75
Antennule	6 segmented, has aesthetasks on 4th 5th, and 6th segments, aesthetask on the 4th segment is the shortest	6 segmented, aesthetask on the 4th segment is the longest, 6th shortest	6 segmented, aesthetask on the 4th segment is the shortest and those of the 5th and 6th segments are equal
Antenna	4 segmented, has accessary structure at the base	4 segmented, devoid of bristles and accessories structure	4 segmented, devoid of accessary structure
Labrum	Bilobed and has sinuate margin	bilobed with deeper notch and sinuate margin	Tetralobed with central two lobes deeply notched sinuate edges
Maxilliped	2 segmented and unornamented	2 segmented, slender and fragile	2 segmented ornamented and devoid of accessary structure
Swimming legs and 5th leg	The basipodites of swimming legs devoid of muscle bands. 5th leg is 2 segmented and the basipodite bears a prominent spine	The basipodites of swimming legs have prominent muscle bands, bears a spine on the basipodite of 5th leg	Prominent muscle band absent. 5th leg devoid of the spine on the basipodite

differences are significant it is proposed to erect a new species and name it *C. bhimilensis.*

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Waltair, *May* 5, 1976.

---

1. Reddiah, K., *J. Zool. Soc. India,* 1961, 12 (2), 137.
2. —, *Crustaceana,* 1961, 2 (4), 300.

An Unusual Record of Oxygen Production in a Freshwater Pond

During the studies (1-10-1974) on the primary production of freshwater ponds of Bhagalpur (India), a very high photosynthetic oxygen production was recorded in one pond known as 'Bhatta Pond'. The 'Light and Dark Bottle' method has been employed for measuring primary production of phytoplankton as described by Gaarder and Gran[6]. Water samples were collected from each 0·5 meter depths with Friedinger sampler and suspended for 24 hours (Nasar and Munshi[10]) at the same depth. Dissolved oxygen was estimated by the unmodified Winkler's method (Ellis *et al.*[5]), and calculated according to Odum[11]. The recorded gross photosynthetic oxygen production (59·00 $gO_2/m^2/day$) in the pond on 1-10-1974 (Table I). appears to be the highest production.

TABLE I
Oxygen production in different water bodies

Water bodies	Gross oxygen production ($gO_2/m^2/$day)	Authors
Refinary holding pond	29·00	Copeland and Dorris[3]
Polluted Estuary	8·70	Hull[7]
Reservoir	5·98	Hull[8]
Sewage pond	26·00	Bartsch[2]
River Lark	35·00	Edwards and Owens[4]
Amaravathy Reservoir	56·91	Sreenivasan[12]
Bhatta Pond	59·00	(Present record)

TABLE II

Physico-chemical properties of Bhatta pond on 1–10–1974

Depth (m) Factors	0·0	0·5	1·0	1·5	2·0
Temperature (°C)	30·2	30·2	29·6	29·6	29·3
Dissolved O_2 (mg/l)	10·3	9·0	7·5	4·2	3·2
Free CO_2 (mg/l)	Nil	Nil	Nil	Nil	Nil
pH	8·2	8·0	7·8	7·8	7·5
Carbonate Alkalinity (mg/l)	7·4	2·1	1·3	1·1	1·1
Bicarbonate Alkalinity (mg/l)	108·0	112·0	128·0	135·0	134·0
Total Hardness (mg/l)	112·0	120·0	120·0	122·0	126·0

On that day a very thick bloom of phytoplankton was visible and distributed even upto one meter. The Secchi disc visibility was 89·0 cm. Bright sunshine was noted for 9 hours on the day. The phytoplankton population was comprised mainly of blue-green algae, which are known to be good photosynthetizers (Nasar[9]). Data on certain physico-chemical properties of the pond water[1] on 1·10·1974, are given in Table II.

The authors are grateful to Professor J. S. Datta Munshi, Dean faculty of Science, Bhagalpur University, for his help and encouragement. Their thanks are also due to the C.S.I.R., New Delhi, for the award of a Research Fellowship.

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1. A.P.H.A., *Standard Methods for the Examination of Water, Sewage and Industrial Wastes, America Public Health Association*, New York, 1965.
2. Bartsch, A. F., *Pymatuning Lab. Field Biol. Spl. Publ. 2*, 1960.
3. Copeland, B. J. and Dorris, T. C., *J. Water Poll. Contr. Fed.*, 1962, 34, 1104.
4. Edwards, R. M. and Owens, M., *Ecology and the Industrial Society*, Blackwell, Oxford 1961, p. 129.
5. Ellis, M. M., Westfall, B. A. and Ellis, E. D., *Res. Rept. 9, Fish and Wild Life Ser.*, 1948.
6. Gaarder, T. and Gran, H. H., *Rapp. Proc. Verb. Conn. Perm. Int. Explor. Mer.*, 1927, 42, 9.
7. Hull, C. J. H., *Low-Flow Augmentation Project*, Rept. No. 14, John Hopkins Univ., 1962, p. 1.
8. —, *Ibid.*, Rept. No. 16, John Hopkins Univ., 1963, p. 1.
9. Nasar, S. A. K., *Ph.D. Thesis*, Bhagalpur Univ., 1975.
10. — and Munshi, J. D., *Japanese J. Ecol.*, 1975, 25, 21.
11. Odum, E. P., *Fundamental of Ecology*, Philadelphia, W. B. Saunders, 1959.
12. Sreenivasan, A., *J. Mar. biol. Assoc., India*, 1975, 7, 1.

Association of *Enterobacter* with Coconut Root (Wilt) Disease

The coconut root (wilt) disease causes an estimated annual loss of about 300 million rupees and has been the subject of multidisciplinary investigations. The symptoms of the disease are a defective root system and flaccidity of leaves and leaflets. Yellowing of leaves and marginal necrosis of leaflets appear during the advanced stages of the disease.

From the growing tips of coconut roots an off white bacterium forming transluscent, smooth, flat and glistening colonies with entire margin was isolated. For isolations, freshly collected and surface sterilized coconut roots from both the root (wilt) affected and healthy regions were plated on an enriched medium containing 15% coconut root extract solidified with agar. The bacterium was conspicuous by its absence in coconut roots collected from the healthy regions.

The bacterium was rod shaped, Gram negative and motile and the cultural characteristics were : utilised glucose fermentatively, catalase positive, MR negative, V.P. positive, H_2S produced, indole negative, gelatin liquefied and starch not hydrolysed. Produced acid and gas from sucrose, **sorbitol**, mannitol, arabinose and salicin besides glucose. Arginine decarboxylase and nitrate reductase were present while lysine decarboxylase, phenyl alanine deaminase and urease were absent. The bacterium has been classified as a species of *Enterobacter* resembling *Enterobacter cloacae.*

The coconut *Enterobacter* isolates produced alcohol precipitable polysaccharide like materials in growing culture filtrates when cultivated as still culture in nutrient broth. An aqueous extract of the polysaccharide like material reversibly wilted tomato seedlings *in vitro.* Species of Aerobacter (*Enterobacter*) have been implicated in plant diseases[1]. Studies are in progress to elucidate the exact role of the bacterium in the coconut root (wilt) disease.

We thank Dr. J. V. Bhat, for critical suggestions, Dr. M. P. Starr, for his untiring efforts on the characterization of the isolates and Dr. J. F. Bradbury, for confirming the identification of the bacterium.

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1. Agrios, G. N., *Plant Pathology,* Academic Press, New York, 1969, p. 328.

Resistant Barley Varieties for the Control of *Heterodera avenae* Wollenweber, 1924

Heterodera avenae, the cereal cyst nematode has a limited favourite host range, *e.g.,* wheat, barley and oat. The disease caused by this nematode is known as 'Molya'. Its distribution[4] in India has been reported from four states including Haryana. According to a report[1], Telone treatment produced a gain in wheat yield (var. Kalyan) to the extent of 128%.

Resistance in barley to *H. avenae* has been reported from abroad[3,5]. Resistant sources were not reported[2] possibly from India. As far as known to the authors, this would be the first report of the kind from this country.

Based upon pot-screening of 75 barley varieties in 1974–75 and testing in 1975–76, a few barley varieties showing resistant reactions in preliminary screening, BP 263, C 164 and B 264 were identified to be highly resistant to *H. avenae.* It was found that ratios of females to males of this nematode in these varieties were 1 : 106, 1 : 119 and 1 : 86 respectively. Thus, formation of enormously larger number of males than females provided a very useful characteristic for heavy reduction in nematode population. A field trial with these varieties has shown that growing of a resistant barley crop in one season in an infested field would check the formation of cysts, and, therefore, almost a normal crop of wheat can be obtained from the same field in the next season. Adoption of this practice is quite effective and simple.

Sub-Department of Nematology,
Haryana Agricultural University,
Hissar, *July* 2, 1976.

D. S. BHATTI.
R. S. DAHIYA.
M. R. DALAL.
S. C. DHAWAN.

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1. Bhatti, D. S. and Dalal, M. R., *HAU. J. Res.,* 1972, 2, 231.
2. Cook, Roggers, *J. Nematol.,* 1974, 6, 165.
3. Cotten, J., *Ann. Appl. Biol.,* 1967, 73, 267.
4. Koshy, P. K. and Swarup, Gopal, *Indian J. Nematol.,* 1971, 1, 106.
5. Nilsson-ehle, H., *Hereditas,* 1920, 1, 1.

Foot Rot of 'Suran' (*Amorphophallus companulatus*)—A New Disease

A severe foot rot of *Amorphophallus companulatus* (Roxb.) Blume ex Dore., was observed (July–September) for the last two years. Isolations revealed the presence of *Fusarium moniliforme* Sheld., from the infected plants.

The disease manifests itself when the plants are about two months old. The rot starts as a water soaked lesion at the collar region. Affected plants show slight pale colour on the tips of young leaves, which gradually spread downwards to the leaf blade and leaf sheath. Ultimately the entire plant turns yellow and collapses due to the rotting of the basal portion. Thiram and Benlate were found effective at 500 ppm. while Captan, Difolatan and Plantvax were in-effective.

Stalk rots due to *Fusarium moniliforme* have been earlier reported on maize by Maric *et al.*[1]. The disease is the first record of *F. moniliforme* causing foot rot of *Amorphophallus companulatus.*

The authors are grateful to Dr. C. Booth, Commonwealth Mycological Institute, Kew, for identification of the fungus and thank Prof. D. D. Pant, Head of the Botany Department, University of Allahabad, for providing necessary facilities.

Department of Botany,
University of Allahabad,
Allahabad 211 002,
March 12, 1976.

S. N. BHARGAVA.
D. N. SHUKLA.
PAVAN KUMAR.

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1. Maric, A., Kosovac, Z. and Jovanov, S., *Agron. Glassn,* 1962, 12, 422.

Note on the Occurrence of *Equisetum hyemale* L. in India

Equisetum hyemale L., previously known to be distributed in many countries is now reported for the first time from India from Kumaon Hills, Do-goan, Nainital (U.P.). The specimens were compared with authentic plants from Jardin Botanico, Rio-de Janiero, Brazil, Central Experimental Farm, Ottawa and pressed herbarium specimens were confirmed by Dr. R. L. Hauke.

The plant was already reported from Northern Asia and W. Asia but not from S. Asia or India. The presence of *Equisetum hyemale* in India is interesting to the pteridophytic flora of India and extends its range of geographic distribution in this part of Asia. The important feature of the distribution is its occurrence in latitude of 28° to 30° North at altitude 2500 to 3100 ft. It is suggested that its occurrence should be explored in the same altitude and latitude particularly in eastern Himalaya.

I am grateful to Dr. Ram Udar and Dr. R. L. Hauke, for their valuable help. Thanks are also due to the Ministry of Agriculture Plant Research Institute, Canada and Director, Jardin Botanico, Rio-de Janeiro, Brazil, for providing the Equisetum specimens.

Department of Botany, DINESH KUMAR SAXENA.
Bareilly College,
Bareilly, India, *June* 14, 1976.

Induction of Polyploidy in Alfalfa

Octoploidy was induced in two varieties of alfalfa (*Medicago sativa* L.), *viz.*, Sirsa-9 and Poona-1-B following colchicine treatment of dry seeds and meristematic tissue of four-day old seedlings. In general, the octoploids had better vigour in terms of height, tillers and leaves but had delayed flowering and seed maturity as compared to the natural tetraploid alfalfa. The size of stomata and pollen grains was increased in the octoploids. Pollen and seed fertility were much reduced in the octoploids. The octoploids may be useful in the production of hexaploid alfalfa for higher forage production[1].

Department of Botany, N. B. KATARE.
Marathwada Agricultural Y. S. NERKAR.
University,
Parbhani (Maharashtra),
May 31, 1976.

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1. Lesins, K., Singh, S. M., Baysal, J. and Sadasivaiah, R. S., *Z. Pflanzenzuchtg.*, 1975, 75, 192.

Initiation of Dikaryon in *Ravenelia esculenta* Naras. and Thirum.

During the course of cytological and developmental studies of the various spore forms of the rust *Ravenelia esculenta*, the authors have made observations on the manner in which the dikaryotization is attained in this fungus.

The seat of dikaryotization is the aecial primordium as in most of the rust fungi. The primary monokaryotic aecial primordium is composed of slightly elongates, rectangular to polygonal cells which are compactly arranged. The initiation of the dikaryon occurs at this stage between adjacent cells by the process of nuclear migration. The migrating nucleus becomes elongated and finds its way into the adjacent cell through a minute pore in the cell wall. After migration the nucleus resumes its original shape and the two nuclei lie side by side. Thus the monokaryotic cells of the aecial primordium become binucleate or dikaryotic.

On the basis of these observations it may be concluded that the process of dikaryotization in *Ravenelia esculenta* is very similar to that of many rust fungi like *Masseeella narasimhanii*[1] and *Puccinia lateritia*[2].

Grateful thanks are due to Dr. G. V. Joshi, for laboratory facilities.

Botany Department, MRS. K. S. HARDIKAR.
Shivaji University, U. K. KULKARNI.
Kolhapur (Maharashtra),
April 20*th*, 1976.

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1. Kulkarni, U. K., *Doctoral Thesis*, University of Poona, 1965.
2. Pawar, I. S., *J. Shivaji Uni.*, 1972, 5(10), 91.

Friedel-Craft Cinnamoylation of Resbenzophenone

During the course of our studies on the Friedel-Craft acylation of resorcinol derivatives, we carried out the cinnamoylation of resbenzophenone. Equimolecular amounts of the latter, cinnamoyl chloride and anhydrous $AlCl_3$ were heated in nitrobenzene solution for 4 hours. After decomposition with dilute HCl followed by steam distillation, the reddish brown pasty residue was washed and triturated with aqueous KOH (10%). The yellow insoluble mass was dissolved in glacial acetic acid and reprecipitated with brine. The product thus obtained crystallised from methanol, m.p. 145–46° (20% yield) and was identified as 5-hydroxy-6-benzoyl flavanone (I). The structure of the latter was confirmed by comparison with the compound prepared by condensing 2-acetyl-4-benzoyl resorcinol (II) with benzaldehyde in the presence of alkali when (I) was directly obtained. The structure of (I) was further confirmed by its bromination in chloroform solution when the corresponding 3-bromoflavanone, m.p. 155°, was obtained as light yellow needles from acetic acid. Reaction of the bromo compound with boiling pyridine afforded the unknown 5-hydroxy-6-benzoyl flavone (III) as colourless needles, m.p. 167–68°. The flavone (III) was also synthesised by the Kostanecki-Robinson benzoylation of 2-acetyl-4-benzoyl resorcinol followed by subsequent treatment with concentrated H_2SO .

All the compounds gave satisfactory analysis for C and H.

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15, Madame Cama Road,
Bombay 400032,
May 6, 1976.

A. S. Gupta.
(late) P. L. Trivedi,
J. R. Merchant.

Efficacy of Colchicine from the New Indian Source

Colchicine has been so long commercially extracted from the corms and seeds of *Colchicum autumnale. C. luteum* and *Merendera* species. Recently a new source has been found in the different members of *Iphigenia* (Liliaceae). The highest yield of colchicine so far recorded is from *Iphigenia stellata* (1·2 to 1·9%)[1,2], whereas the former species of Liliaceae yield 0·5 to 0·21% only[3].

Colchicine extracted from the new source has been applied on *Allium cepa* to test its efficacy as polyploidizing agent. The results are very promising. Cytological effects like metaphase arrest, different degree of polyploidy, multipolarity, diplochromatids, etc., are very predominent with all the three concentrations applied. The concentrations used are 0·01, 0·1 and 0·5% respectively.

The results have been compared with similar concentrations prepared from E. Merck sample. *Tumours* were induced on the root tips after 24 hours of treatment (0·01%). The divisional frequecy with the above concentration from the new colchicine ranges from 8–30% whereas with that of E. Merck colchicine, the frequency is 1–23° .

The small stock of colchicine used in this experiment was obtained through courtesy of Dr. R. S. Rao, Deputy Director, Central National Herbarium, Botanical Survey of India.

Cytology Section,
Central Botanical Laboratory,
Botanical Survey of India,
Howrah 3, *June* 11, 1976.

R. Mallick.
A. K. Sarkar.

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1. Kapadia, V. H., Sukh Dev, Rao, R. S. and Ansari, M. Y., *Phytochemistry*, 1972, **11**, 1193.
2. Ansari, M. Y. and Rolla, S. Rao, *Bull. Bot. Surv. India*, 1976, **15**, 118.
3. Eigsti, O. J. and Dustin, P. Jr., *Colchicine in Agriculture, Biology, Medicine and Chemistory*, Iowa State College Press, U.S.A., 1957.

REVIEWS AND NOTICES OF BOOKS

Matrices and Tensors in Physics. By. A. W. Joshi. (Wiley Eastern Ltd., New Delhi), 1975. Pp. x + 250. Price Rs. 15·00.

This is a good book, welcome to the stock of texts to be used by the physics students at the University B.Sc. and M.Sc. level. The subject of matrices appears first in connection with the solution of simultaneous algebraic equations and then permeates into other areas especially quantum mechanics. Tensors, restricted to cartesian tensors, are applicable at an intermediate level to a number of areas in elasticity, crystal physics and so on, whereas the full blast of tensor analysis becomes indispensible when dealing with general relativity. The book makes a sincere attempt to introduce these two topics for the students and should succeed in its task.

The coverage of the topics is fairly conventional. On the first topic, one starts with the algebra of matrices, special types of matrices, solutions of simultaneous equations, diagonalization and eigenvalue-eigenvector problems and finally deals with functions of matrices. On the second topic one starts with the definition and algebra of tensors, the metric tensor, tensor calculus and Christoffel symbols and goes to a discussion of the curvature tensor.

What distinguishes the book is the presence of about 100 worked examples and about 200 problems with partial answers, selected from different areas of physics. This should drive home the point that the subject is not an isolated study but is applicable to may areas of physics. The book bears the mark of having been taught earlier to groups of students. In a sense it is a tested product which can be recommended. The book, published with a subsidy from the National Book Trust, is also moderately priced at Rs. 15/– and deserves to be popular among the students.

E. S. R. Gopal.

Drought and Agriculture. By C. E. Hounam, J. J. Burgos, M. S. Kalik, W. C. Palmer and J. Rodda. (Technical Note No. 138, published by the World Meteorological Organization, Geneva, Switzerland), 1955. Pp. i to xv + 127. Price not given.

This is the report of the working group on the assessment of drought which was set up by the World Meteorological Organization with Mr. C. E. Hounam as the Chairman and Messrs. J. J. Burgos, M. S. Kalik, W. C. Palmer and J. Rodda as Members.

This technical note gives a systematic analysis of the drought problem from the agricultural point of view. The various definitions of drought classified according to the criteria such as rainfall, rainfall with mean temperature, soil water and crop parameters, climatic indices and estimates of evaporation, and other general definitions have been given. The different meteorological indices of agricultural drought, methods of analysis of the rainfall data and water balance equations developed by the use of climatic parameters have been explained in detail. Nature of drought injury to plants, structure and mechanical features of plants in relation to drought resistance, the behaviour of plant communities as in pastures and the effect of drought on livestock and water requirements in general are also presented.

Drought conditions lead to direct losses from reduced crop yields, pasture deterioration and at times death of livestock. There are number of indirect losses affecting the socio-economic conditions of the people. As such understanding the drought and planning to mitigate the drought effects become necessary. This technical note which contains the recent research work can be made use of for planning development of drought-prone areas. This publication is greatly useful to the research workers and for those involved in agricultural planning.

G. V. Havanagi.

Mulching Effects on Plant Climate and Yield. By J. W. Davies. (Technical Note No. 136, published by the Secretariat of the World Meteorological Organization, Geneva, Switzerland), 1975. Pp. i to xi + 92. Price not given.

The Commission for Agricultural Meteorology set up by the World Meteorological Organization discussed the topic of importance of mulches on the successful production of crops at the Fifth Session of the Commission held in 1971 and Mr. J. W. Davies was the rapporteur on Meteorological Effects of soil cover. This publication contains the report.

The review of published papers between 1954 to 1970 concerning the effect of mulches on crop yields and micro climate of the soil are included. Effects of mulches on temperature, soil moisture, erosion and soil physics, pests and diseases, growth and yield of crops and weed suppression are the major topics presented. Each part is subdivided into no specific crop, temperate fruit trees, temperate soft fruits, vegetable and flowers, forestry, grasses and cereals and tropical and sub-tropical crops. Effect of the organic and inorganic mulches are reviewed separately and mixed mulch comparisons are also made wherever needed.

Mulching or covering of the soil surface with organic or inorganic mulches help in decreasing the loss of soil moisture due to evaporation, reducing the soil temperature fluctuations and protecting the soil from the impact of rain thereby minimising the soil loss due to erosion. The mulches can be used advantageously for the cash crops or fruit trees wherever they are grown under limited rainfall or irrigation water.

This publication is very useful as a reference book for workers in agriculture involved in research on the efficient use of soil moisture.

G. V. Havanagi.

ANNOUNCEMENTS

Award of Research Degrees

Karnatak University, Dharwar, has awarded the Ph.D. degree in Physics to Shri Patil Ranganagoud Laxmanagoud ; Ph.D. degree in Chemistry to Shri Narke Chandrashekhar Sharanappa ; Ph.D. degree in Botany to Shri Sadanand Nagesh Hegde.

M.S. University of Baroda has awarded the Ph.D. degree in Chemistry to Kumari Jasoda Hariram Pardanani ; Ph.D. degree in Biochemistry to Shri Hira Lal Nakhasi ; Ph.D. degree in Zoology to Shri Pratap Lalsing Yadav ; Ph.D. degree in Microbiology to Shri Ashutosh Nilkanthrai Kachhy ; and Kumari Salma Mohammed Amin Baqui ; Ph.D. degree in Physics to Shri Arvindkumar Ramanlal Vyas.

Osmania University, Hyderabad, has awarded the Ph.D. degree in Biochemistry to Shri S. Bapu Rao ; Ph.D. degree in Chemistry to Shri Satyanarayana Murthy ; Ph.D. degree in Physics to Shri .E. Narasimhacharyulu ; Ph.D. degree in Genetics to Smt. L. Geetha.

Sri Venkateswara University, Tirupati, has awarded the Ph.D. degrees in Botany to Shri T. Haragopal ; to Shri M. Narasimha Reddy ; and to Shrimathi M. Santha Kumari ; Ph.D. degree in Physics to Shri N. Manohara Murthy.

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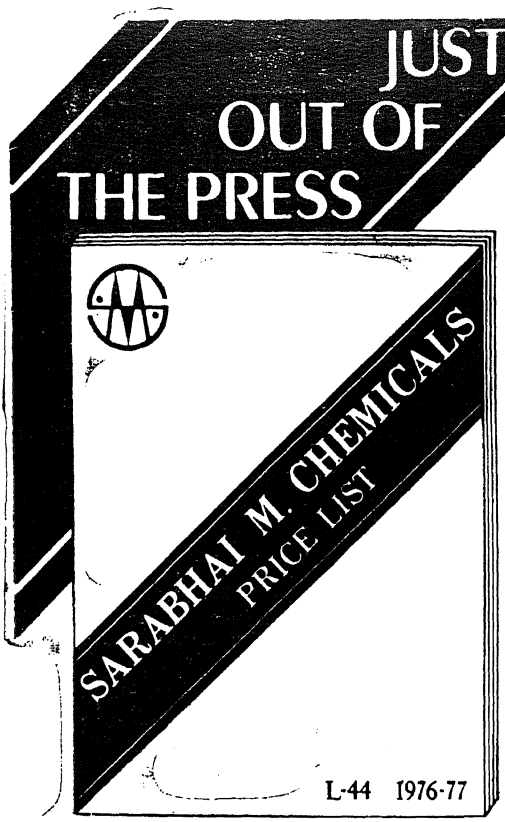

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THE BANGALORE PRE

PROTON MAGNETIC RESONANCE STUDY OF SOLID L-GLUTAMIC ACID AT ROOM TEMPERATURE

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THE present study of l-glutamic acid is oriented towards the check of the crystal structure[1] and making sure the Zwitterion Structure at room temperature as modified by Hirokawa.

CRYSTAL DATA AND HYDROGEN BONDING IN L-GLUTAMIC ACID

Bernal[2] reported the crystal structure of l-glutamic acid to have cell dimensions $a = 7{\cdot}06$; $b = 10{\cdot}3$ and $c = 8{\cdot}75$ Å, space group $P2_12_12_1$ and $z = 4$ orthorhombic in form. E.D.P. studies of Hirokawa[1] presented a new modification of the structure in which cell dimensions $a = 5{\cdot}7$, $b = 17{\cdot}34$ and $c = 6{\cdot}95$ Å and $z = 4$ with space group $P2_12_12_1$ and orthorhombic in form. The infrared studies[3-5] associate a zwitterion structure to l-glutamic acid. The cyanoethylation and microwave studies[6,7] also show the zwitterion properties of this compound. The carbon chain configuration is not coplanar and the structure consists of molecules tied into infinite chain along the direction of the *b*-axis by O–H···O hydrogen bonds and these chains are linked together in other direction by N–H..O hydrogen bond. In the same molecule H atom is located at a distance of 1·09, 1·01 and ·97 Å from C, N and O atoms respectively. The distance between two oxygen atoms of the two carboxyl groups in the molecule is 2·54 Å. The structure of l-glutamic acid gives information regarding the atomic arrangement in a polypeptide chain and the way in which these chains occur in natural proteins. The structure of the chain folded poly (l-glutamic acid)[8] single crystals accounts for the principal features of the chain.

It is concluded[9] that in the study of the conformational analysis of solid amino acids, the variations in the bond lengths and bond angles can be nearly neglected. The structure of l-glutamic acid is an example of the fact that while the bond distances and valency angles in the carbon chain of different amino acids remain approximately unchanged, each amino acid assumes a different configuration with respect to a particular C–C bond. The hydrogen bonds play an important role in the configuration of the polypeptide chain and its relation to neighbouring atom. In solutions l-glutamic acid occurs in helical conformation[10,11].

EXPERIMENTAL DETAILS

The NMR spectrum of the high purity sample of l-gutamic acid was recorded at room temperature with a 12 inch magnet system at a resonance frequency of 7·5 Mc/S at T.I.F.R. (Bombay). The details of the spectrometer are given elsewhere[12].

CALCULATIONS

Theoretical Second Moment.—There are two contributions to rigid lattice second moment, the S_1, called intra-molecular, arising due to the interaction between the protons of the same molecule and, S_2, the inter-molecular, arising due to the interaction between the protons of the different molecules situated at the various lattice sites. The evaluation of S_1 is made by the Van Vleck's formula[13] for the powder sample.

$$S_1 = \frac{716{\cdot}55}{N} \sum_{j>k} r_{jk}^{-6} \qquad \text{(i)}$$

where N is the number of protons in the molecule and r_{jk} is the inter-nuclear distance between the *j*th and *k*th nuclei. The modified molecular model of Hirokawa[1] was used according to which cell dimensions are $a = 5{\cdot}7$; $b = 17{\cdot}34$ and $c = 6{\cdot}95$ Å and $z = 4$, with space group $P2_12_12_1$ and orthorhombic in form. The value of S_1 was computed to be 11·99 gauss^2.

The inter-molecular contribution S_2 to rigid lattice second moment arises out of the interaction between the protons of the different molecules deposited at the various lattice sites and exact evaluation of this contribution is too tedious. An approximate estimation can be made following the method of Andrew and Eades[14] and the value of S_2 was found to be 6·00 gauss^2. This value appears to be quite reasonable as Agarwal *et al.*[15] obtained this value 6·3 gauss^2 in case of solid L-alanine, a similar compound. At this point it can be mentioned that further check of the value of S_2 can be made by the formula of Smith[16]. Thus the value of the theoretical rigid lattice second moment is then $11{\cdot}99 + 6{\cdot}00 = 17{\cdot}99$ gauss^2.

Experimental Second Moment.—The experimental value of the second moment are calculated from the NMR line shape derivative $g\,(\mathrm{H})$ using expression

$$S = \tfrac{1}{3}\left[\frac{\int_0^\infty g\,(\mathrm{H})\,(\mathrm{H}-\mathrm{H}_0)^3\,d\mathrm{H}}{\int_0^\infty g\,(\mathrm{H})\,(\mathrm{H}-\mathrm{H}_0)\,d\mathrm{H}}\right] - \tfrac{1}{4}\,\mathrm{H}^2 m \qquad \text{(ii}$$

where H_0 is the value of field at resonance and H*m* is sinusoidal field modulation amplitude. The second term in the expression occurs due to suggestions of Andrew[17]. The accuracy in the measurement is about $\pm 1{\cdot}0$ gauss^2. The experimental second moment at room temperature comes out to be 16·3 gauss^2. Due to experimental difficulty, investigations of the sample at different temperatures could not be carried out.

RESULTS AND DISCUSSION

The observed value of second moment at room temperature is 16·3 gauss^2. The theoretically computed value of second moment for the rigid lattice is 17·99 gauss^2. A reasonable agreement exists in the theoretical and experimental value of second moment within the extent of experimental error. The lattice is rigid at the room temperature. The conformity between our experimental and the theoretical values of second moment justify the E.D.P. structure[1].

There exists no evidence of any sort of motion in the solid *l*-glutamic acid by electron density projection study or any other study at room temperature. This seems reasonable. The probability of molecular reorientation is least because of the relatively large size of the molecule and the way in which molecules link[1]. The asymmetric carbon atom of the molecule is linked to the amino group, the $(CH_2)_2$ COOH group and the carboxylic group. The rotation of $(CH_2)_2$ COOH group is also not feasible about the C–C bond for the reasons of its mass and being attached to N atom which is surrounded by three oxygen atoms through at least two hydrogen atoms and the rotation of this group will involve the disturbance of extreme O atom separation of 2·54 Å. Similarly *r*-carboxylic group too, cannot rotate at a low temperature due to its structural features[18]. Again the amino group is linked to the other molecule by N–H···O bond and requires sufficient energy to reorient, depending upon the temperature[19–21] of the transition. The evidence of amino group reorientation at high temperatures in solid amino acids are numerous[15,22] and the problem of rotating interaction in connection with the fall of second moment value is dealt tacitly by Smith[23] and Kromhout *et al.*[24]. Thus in our case the amino group rotation may take place at higher temperatures where the hindrance offered due to hydrogen bonding between the different substituent groups and also between different molecule is overcome. It is to be noticed that hyperfine structure of E.S.R. spectrum[25,26] due to interaction of free electron spin with the hydrogen nuclei and the irreversible changes in the E.P.R. spectra[27,28] at high temperatures and fine structure of P.M.R. derivative trace which is a consequence of the presence of the chemically non-equivalent protons; all are informative of the reorientation of the amino group at high temperatures. Again some of the probability of CH_2 group and the carboxylic group (not the *r* one) rotation seems to exist from microwave[7] magneto-chemical[29] and U.V. absorption studies[30] but the structural features permit none of them at room temperature and a conclusive character of group reorientation can be ascribed only after the study at different temperatures and the respective second moment variations, which we could not study due to experimental difficulty.

ACKNOWLEDGEMENT

We sincerely thank Prof. R. Vijairaghavan of T.I.F.R. (Bombay) for the experimental facilities and Dr. B. G. Gokhale, Head of the Department of Physics, for his keen interest and encouragement.

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1. Sakutaro Hirokawa, *Acta Cryst.*, 1955, **8**, 637.
2. Bernal, J. D., *Z. Kristallogr.*, 1931, **78**, 363.
3. F. William, R. and M. John, T., *Appl. Spectros.*, 1971, **25** (2), 175.
4. Balueva, G. R. and Terskov, I. A., *Sb. Dokl. Sib. Soves, Spektrosk. 3rd Krasnoyarsk USSR*, 1964, p. 140.
5. Patricia, N. Wancheck and Jeantte, G. Grassell, *Appl. Spectros.*, 1967, **21** (5), 336.
6. Mc Kimey, L. L., Uhing, E. H., Setzkorn, E. A. and Cowan, J. C., *J. Am. Chem. Soc.*, 1950, **72**, 2599.
7. Walter, G. William, B. and Hoeards, S., *Proc. Natl. Acd. Sc. U.S.*, 1955, **41**, 983.
8. Keith Harvey, D., Padden Frank, J. and Giannoni, G., *J. Mol. Bio.*, 1969, **43** (3), 423.
9. Lakshminarayanan, A. V., Sasisekharan, V. and Ramachandran, G. N., *Conform. Biopolym. Pap. Int. Symp. (Mad.)*, 1967, **1**, 62.
10. Brahms, J. and Spach, G., *Nature*, 1963, **200** (4901), 72
11. Akiyoshi, Wada, *J. Chem. Phys.*, 1959, **30**, 328.
12. Agarwal, V. D., *NMR Study of Some Organic Compounds*, Ph.D. Thesis, Lucknow University, Lucknow, 1970.
13. Van Vleck, J. H., *Phys. Rev.*, 1948, **74**, 1168.
14. Andrew, E. R. and Eades, R. G., *Proc. Phys. Soc.*, 1953, **A66**, 415.
15. Agarwal, S. C., Agarwal, P. and Gupta, R. C., *Journ. f. Prat. Chemie.*, Heft 3, 1973, S. 443.
16. Smith, G. W., *J. Chem. Phys.*, 1965, **42**, 4299.
17. Andrew, E. R., *Phys. Rev.*, 1953, **91**, 425.
18. Doyne, T. M., Hirokawa, S. and Watanabe, C., *Acta Cryst.*, 1954, **7**, 652.
19. Gutowsky, H. S. and Pake, G. E., *J. Chem. Phys*, 1950, **18**, 162.
20. Kubo, R. and Tomita, K., *J. Phys. Soc. Japan*, 1954, **9**, 888.
21. Waugh, J. S. and Fedin, E. I., *Fiz. tverdogo tela*, 1963, **4**, 1633.
22. Banerjee, A. K., Mirza, P., Agarwal, P. A. and Gupta, R. C., *Ind. Jour. of Pure and Appl. Phys.*, 1974, **112**, 768.
23. Smith, G. W., *J. Chem. Phys.*, 1965, **42**, 4229.
24. Kromhout, R. A. and Moulton, W. G., *Ibid.*, 1955, **23**, 1673.
25. Mc Cosmic Gen, G. and Gordy, W., *J. Phys. Chem.*, 1958, **62**, 783.
26. Russel, C. Drew and Gordy, W., *Radiation Res.*, 1963, **18** (4), 552.
27. Mochal Rion, A. I. and Kharitonenkov, I. G., *Tr. Mosk. Obsochest. Ispyt. Priorotd. Bio.*, 1966, **16**, 113.
28. Albert Van de Vorst, *C.R. Acad. Sci.* (Paris), 1967, Ser, D, **264** (8), 1100.
29. Roland Per Cean, *Compt. Rend.*, 1953, **236**, 76.
30. Vinogradov, I. P., *Dodonova Nya. Opt. Specktrosk*, 1971, **30** (1), 27.

IONOSPHERIC SCINTILLATIONS INDUCED BY CLOUD OF INTENSE SPORADIC E LAYER

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ABSTRACT

The simultaneous observations of vertical ionospheric soundings and the faraday fadings of the beacons from a low orbiting satellite have shown clearly that the existence of a cloud of intense sporadic E in the path of radio waves does produce scintillations even during the day time at low latitudes. The latitudinal width of E_s cloud is estimated to be around 300 km.

INTRODUCTION

EARLY studies of ionospheric scintillations were carried out at middle and high latitudes using the signals from radio stars which traversed the whole ionosphere. The signals received were found to fade violently during the night time. This phenomenon known as ionospheric scintillation of radio waves was found to be associated with the occurrence of spread F in the path of radio waves (Wild and Roberts, 1956; Booker, 1958).

With the advent of artificial earth satellites, scintillation studies have been extended to all latitudes. In middle latitudes again, a positive correlation was found between scintillations and spread F (Yeh and

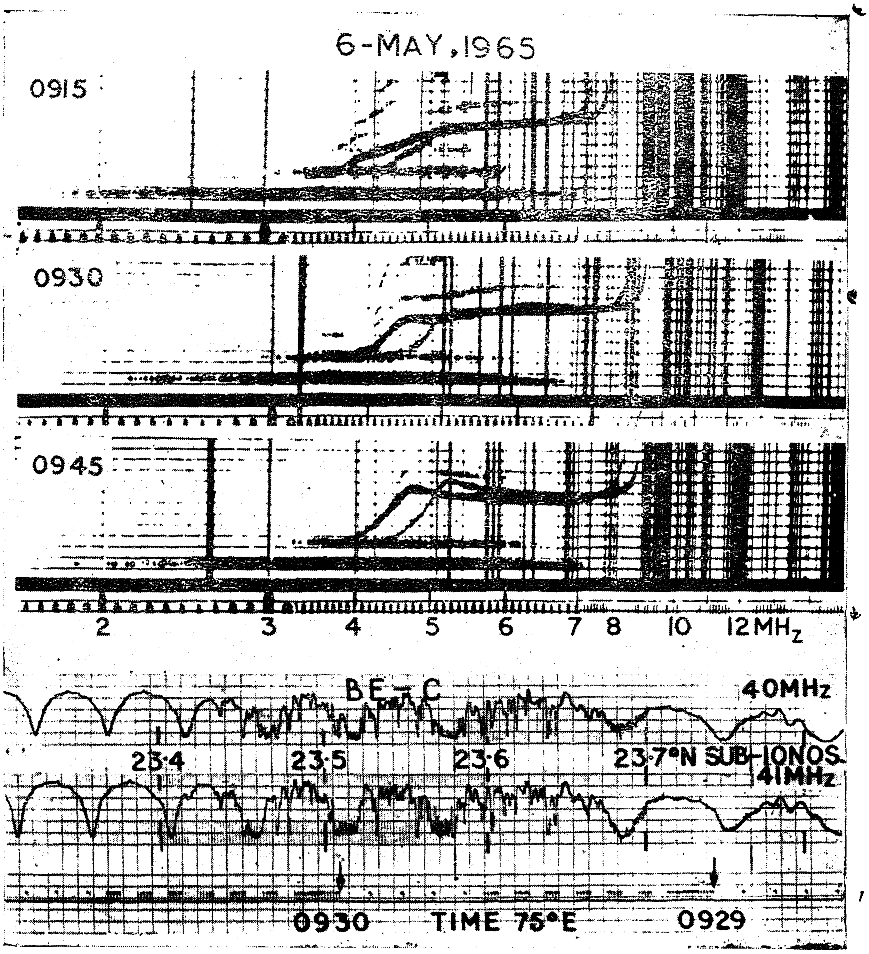

FIG. 1. Faraday fading records at 40 and 41 MHz from the BE-C Satellite recorded at Ahmedabad on 6 May 1965 showing scintillations (bottom), caused by the intense sporadic E seen in the ionograms at the top.

Swenson, 1959; Beynon and Jones, 1964; McClure, 1964). At equatorial latitudes too, the scintillations were found to be a night time phenomenon associated with spread F (Kent, 1961; Koster, 1972; Chandra and Rastogi, 1974).

At high latitude stations scintillations have been found to occur some times during the day and are attributed to E-region irregularities (Bolton *et al.*, 1953; Dueno, 1956; Chivers and Greenhow, 1959; Munro, 1966). Using spaced receivers McClure (1964) estimated the height of irregularities causing the day time scintillations to be at 100 km. Aarons and Whitney (1968) showed that the day time scintillations observed at Sagamore Hill (dip 73° N) were correlated with the occurrence of E_s, the width of E_s cloud estimated by them was 300–600 km. At a low latitude station, Brisbane (20° N), Singleton and Lynch (1962) did not find any significant correlation between scintillations and f_0E_s. Koster and Wright (1963) have observed an unusually regular type of scintillation during day time at Accra (dip 8·5° S) which was associated with equatorial electrojet disturbances. Ramakrishnan (1966) has reported the associations between the occurrences of scintillations and E_s. The present note is concerned with a conclusive evidence for stroeng blanketing type of E_s causing scintillations at 40 and 41 MHz.

RESULTS

In Fig. 1 are shown the ionograms taken at Ahmedabad at 0915, 0930 and 0945 hr 75° EMT on 6 May 1965. All these records show a strong E_s cloud over Ahmedabad with $fE_s > 7 \cdot 0$ MHz, and the second multiple of E_s is also clearly seen to be strong. In the lower part of the diagram are shown the fading records of 40 and 41 MHz radio beacons from the Satellite BE–C recorded at Ahmedabad. The sub-satellite pass was very close to the location of Ahmedabad, the maximum elevation of the satellite was 82° at 0930 hr 75° EMT. The fading records for periods prior to 0929 hr, *i.e.*, for sub-ionospheric position north of 23·7° N showed very clear Faraday fades. Between 09*h*, 29′, 12″ and 09*h*, 30′ 20″ strong scintillations were superimposed over the Faraday fades. The sub-ionospheric latitude varied from 23·7° N to 23·4° N during this period. After 0931 hr, when the satellite was south of 23·4° N the Faraday fades were again quite clear. This showed that strong irregularities were present in the ionospheric between latitues of 23·4° N and 23·7° N. This clearly shows a distinct correlation of strong blanketing E_s and the scintillations produced in the radio waves traversing through it. The E_s cloud seems to be about 300 Km wide in N-S direction.

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1. Aarons, J and Whitney, H. E., *Planet. Space Sci.*, 1968, **16**, 21.
2. Beynon, W. J. G. and Jones, E. S. O., *J. Atmos. Terr. Phys.*, 1964, **26**, 1175.
3. Bolton, J. G., Slee, O. B. and Stanley, G. J., *Australian J. Phys.*, 1953, **6**, 434.
4. Booker, H. G., *Proc. IRE.*, 1958, **46**, 298.
5. Chandra, H. and Rastogi, R. G., *Curr. Sci.*, 1974, **43**, 567.
6. Chivers, H. J. A. and Greenhow, J. S., *J. Atmos. Terr. Phys.*, 1959, **17**, 1.
7. Dueno, B., *J. Geophys. Res.*, 1956, **61**, 535.
8. Kent, G. S., *J. Atmos. Terr. Phys.*, 1961, **16**, 10.
9. Koster, J. R., *Planet. Space Sci.*, 1972, **20**, 1999.
10. Koster, J. R. and Wright, R. W., *Radio Astronomical and Satellite Studies of the Atmosphere*, Edited by J. Aarons, North Holland, Amsterdam, 1963, p. 114.
11. McClure, J. P., *J. Geophys. Res.*, 1964, **69**, 2774.
12. Munro, H., *Rad. Sci.*, 1966, **1**, 1186.
13. Ramakrishnan, S., *Ph.D. Thesis*, Kerala University, 1966.
14. Singleton, D. G. and Lynch, G. J. E., *J. Atmos. Terr. Phys.*, 1962, **24**, 353.
15. Wild, J. P. and Roberts, J. A., *J. Atmos. Terr. Phys.*, 1956, **8**, 55.
16. Yeh, K. C. and Swenson, G. W., *J. Geophys. Res.*, 1959, **64**, 2281.

PREPARATION AND CHARACTERISATION OF ORTHOPHENYLENEDIAMINE COMPLEXES OF LANTHANIDES

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ORTHOPHENYLENEDIAMINE (OPDA) complexes of Fe (II) halides were prepared and characterised by infrared, electronic, Mössbauer spectroscopy and magnetic susceptibility measurements[1]. Some complexes of Co (II), Ni (II) and Cu (II) with orthophenylenediamine have been prepared and characterized by magnetic and spectral properties[2]. In all the above complexes it was observed OPDA acts either as a bidentate ligand or as a unidentate ligand.

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No information has been reported on lanthanide complexes except a spectrophotometric determination of Ce (IV) with OPDA. This communication deals with the preparation and characterization of Y, La, Pr, Nd, Sm and Gd complexes with OPDA. The rare earth OPDA complexes were prepared adopting the procedure of Rao *et al.*[3], who slightly modified the procedure adopted by Sacconi and Ercoli[4] and Moeller[5]. All the solid complexes obtained were washed with ether and vacuum dried for 48 hours and analysed for metal, nitrogen and anion. The analytical data indicated that each lanthanide ion was attached to three moles of OPDA. A study of the conductivity data indicated that all the complexes are 1 : 3 electrolytes. The analytical data of the several lanthanide OPDA complexes is detailed in Table I.

In the neodymium complex, only two bands are located at 590 and 630 nm. Here also it is likely that the 576 nm characteristic band has undergone a red shift and splitting also. Such a splitting was observed by Moeller[6] in a study of Nd–EDTA complex. The characteristic 402 nm band of the Sm^{3+} ion could not be located in the complex.

Orthophenylenediamine can function as a bidentate or unidentate ligand in metal complexes. By a study of the infrared spectra of Cu (II), Cd (II), Co (II), Ni (II), Zn (II)[7,8] complexes it was reported that one could distinguish between the chelated and unidentate situation of the ligand. The type of bonding in each complex was determined by using N–H and C–N stretch frequencies as indicators.

TABLE I

Complex	% Metal		% Chloride		% Nitrogen	
	Calc.	Obsd.	Calc.	Obsd.	Calc.	Obsd.
Y (OPDA)$_3$ Cl$_3$H$_2$O	16·53	16·36	19·80	19·39	15·63	15·15
La (OPDA)$_3$ Cl$_3$H$_2$O	23·63	23·22	18·11	17·79	14·29	14·05
Pr (OPDA)$_3$ Cl$_3$H$_2$O	23·88	23·35	18·05	17·95	14·24	14·10
Nd (OPDA)$_3$ Cl$_3$H$_2$O	24·31	24·18	17·95	17·54	14·18	13·92
Sm (OPDA)$_3$ Cl$_3$H$_2$O	25·08	24·77	17·77	17·27	14·02	14·01
Gd (OPDA)$_3$ Cl$_3$H$_2$O	25·93	25·89	17·57	17·20	13·86	13·29

RESULTS AND DISCUSSION

In this investigation the ligand OPDA has shown three bands in the ultraviolet region at 210, 240 (*sh*) and 295 nm. In the lanthanide complexes studied, the 210 nm band is located in all of them except in the gadolinium complex. In the gadolinium complex only the 240 nm (*sh*) has been located. However, in the case of La, Pr and Nd complexes the 240 (*sh*) and the 295 nm bands have merged with each other and a distinct band at 260 nm is located. In the samarium complex two bands at 235 and 290 nm are located. There is an enhancement in the log ϵ values in the complexes at the reported maxima in comparison with the value of the free ligand. The log ϵ values are of the order 3·94–4·94. These changes can be taken as an involvement of the nitrogens of the ligand in bond formation.

In the praseodymium complex the 444 nm characteristic band of the praseodymium ion is observed at 450nm and two bands at 580 and 630 nm have been observed. Probably the 590 nm characteristic band of the praseodymium ion has been split into two inflections. However, the 480 nm characteristic band could not be observed.

In the orthophenylenediamine investigated[1], two bands are observed at 3365 and 3340 cm^{-1}. It was postulated that the free amine groups show bands above 3350 cm^{-1} while the coordinating groups have no strong bands above about 3300 cm^{-1}.

In this investigation the ligand has shown two bands at 3370 and 3350 cm^{-1} which can be assigned to N–H stretch. In all the complexes investigated there is a band around 3385 cm^{-1}. A band around 3305 cm^{-1} was located in La, Pr, Nd and Sm complexes. It can be said that the 3370 cm^{-1} band is located with slight shift whereas the 3350 cm^{-1} band has disappeared in Y and Gd complexes and lowered in the rest of the complexes and the lowering is of the order of 45 cm^{-1}. From the changes observed in the N-H stretch frequency in the lanthanide complexes, one can postulate as a first approximation that OPDA possibly acted as a bidentate ligand.

In this investigation, in the free ligand there are two bands at 1273 and 1248 cm^{-1} which are due to C–N stretch. The intensity of these bands can also be qualitatively taken as a measure for determining the nature of bonding of the ligand. If the nitrogens are involved in bonding, there is naturally a decrease in intensity. In the lanthanide complexes now investigated, both the 1273 and 1248 cm^{-1} bands of the ligand have been affected in position and there is considerable decrease in intensity.

There is broad band between 3500–3600 cm^{-1} in all the lanthanide complexes and this band is due to the presence of water molecules in the complexes. However, there is no band in the free ligand in this region.

From an examination of the above data, it can be concluded that orthophenylenediamine acted as a bidentate ligand in the lanthanide complexes.

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1. Renovitch, G. A. and Baker, W. A. (Jr.), *J. Chem. Soc.* (A) (1) **75** (1969).
2. Duff, E. J., *J. Chem. Soc.*, 1968, A434.
3. Purushotham, D., Ramachandra Rao, V. and Raghava Rao, Bh. S. V., *Anal. Chim. Acta*, 1965, **32**, 182.
4. Sacconi, L. and Ercoli, R., *Gaz. Chim. Ital.*, 1949, p. 731.
5. Moeller, T. and Ulrich, W. F., *J. Inorg. Nucl. Chem.*, 1956, **2**, 164.
6. — and Bradley, J. C., *J. Am. Chem. Soc.*, 1950, **72**, 5447.
7. Heifer, W., Schlissmann, C. and Ries, K., *Z. Anorg. Chem.*, 1929, **180**, 89.
8. Barvinok, M. S. and Bhukareva, I. S., *Zur. Neorg. Khim.*, 1965, **10**, 861.

STUDIES ON *COSTUS SPECIOSUS* SIMS.

Part I. Variability in Diosgenin Content

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Abstract

A wide range of variation in the diosgenin content has been observed in the rhizomes of *Costus speciosus*. The distribution in different portions of the same rhizome is uneven. There is a definite clone to clone variation while the diosgenin content is highest during the full bloom period in the months of July and August.

Introduction

COSTUS SPECIOSUS SIMS. has recently gained importance as a source of diosgenin, a precursor in the synthesis of steroidal hormones[1]. Analysis of a large number of rhizome samples collected from various localities during different months revealed a lot of variation in diosgenin content from plant to plant as well as at different periods of harvest. Detailed studies were, therefore, undertaken to find out the range of variability in the diosgenin content and the factors affecting it.

Materials and Methols

Four plants from one year old crop in the nursery were harvested during December, 1975 for determining the distribution of diosgenin in different portions of the same rhizome. Each rhizome was divided into tips, nodes and internodes and each set analysed separately for diosgenin content. For determining the range of diosgenin content in the natural population, two hundred plants growning side by side in the Suhari Village were harvested during November, 1975 and each of the rhizome analysed separately. Determination of the diosgenin content during different months of the year was done by sampling three plants from a two year crop in the nursery. A uniform portion of the rhizome of each plant was taken for analysis at 50 days interval between January, 1975, the period when the dormancy sets into April, 1976 when buds on the nodes start swelling. The procedure adopted for chemical analysis is according to Sarin *et al.* (*loc. cit.*). The diosgenin content has been calculated on dry weight basis.

Results and Discussions

(i) The diosgenin content in tips, nodes and internodal portions of the rhizome are given in Table I. The diosgenin content in the nodal regions is greater than the tips and internodes, the latter having the least amount. The lesser concentration of diosgenin in tip and internodal regions, however, does not affect much the overall yield from the rhizome as the bulk of its biomass (about 75%) is formed by close set nodes.

(ii) *Range of diosgenin content in natural population*: The analysis of 200 rhizome samples indicate that the diosgenin calculated on crude basis range from 0·39% to 3·65% (d.w.b.). Of the seven categories of clones containing between 0·5% and 4·0% of diosgenin, those having upto 1% constitute only 2% of the total, while those containing 1·0% to 1·5%, 1·5% to 2%, 2·5% to 3·0%, 3·0% to 3·5% and 3·5%

TABLE I

Distribution of diosgenin in one year old rhizome of Costus speciosus

Diosgenin percentage (Pure: dry wt. basis)

Tuber portion	Sample No. 1		Sample No. 2		Sample No. 3	
	diosgenin %	Increase or decrease over average	diosgenin %	Increase or decrease over average	diosgenin %	Increase or decrease over average
Nodes with sprouted stem	0·88	+0·27	0·57	+0·14	0·72	+0·15
Internodes	0·41	−0·20	0·36	−0·07	0·42	−0·15
Tips	0·60	−0·01	0·37	−0·06	0·56	−0·01
Average	0·61	..	0·43	..	0·57	..

TABLE II

Seasonal variation in diosgenin content of Costus speciosus *rhizome*

Date of sampling	Vegetative stage	% Diosgenin (Pure: dry wt. basis)		
		Clone (1)	Clone (2)	Clone (3)
1st January, 1975	Aerial portions dried up	0·63	0·52	0·61
20th February, 1975	Dormant	0·91	0·75	0·86
10th April, 1975	Dormant	0·80	0·81	0·81
30th May, 1975	Aerial shoots 10–15 cm tall with few leaves	0·65	0·69	0·65
21st July, 1975	Aerial shoots with flower buds	2·61	2·02	2·64
10th September, 1975	Fruit formation on lower flowers of the inflorescence	1·02	0·98	0·88
30th October, 1975	Flower heads with mature fruits lower leaves yellowing	1·21	0·86	0·34
20th December, 1975	Aerial portions dried up	0·72	0·38	0·44
8th February, 1975	Dormant	1·03	0·45	0·50
30th March, 1976	Dormant	1·10	0·69	0·61

to 4% of diosgenin constitute 14%, 30%, 39%, 12%, 2·5% and 0·5% of the total respectively. It can be noticed that more than half of the total number of clones sampled (54%) contain more than 2% of crude diosgenin. Somewhat similar observations have been made in certain other localities in Jammu, Kangra and Dehradun. The fact that some of the clones are exceptionally rich in diosgenin, promises great possibilities of evolving a high yielding strain through selection and multiplication.

(iii) *Seasonal variation in diosgenin content*: The analysis of rhizome portions obtained from three plants have revealed interesting results (Table II). The diosgenin content increases steadily from early January when complete dormancy sets in till the month of May when young shoots sprout. There is a very sharp increase in the months of July and August when the plants achieve optimum height and are in full bloom. Diosgenin content again declines sharply during September and October, the period of intensive fruit formation. This decline continues till the last week of December when the aerial shoots dry up completely. The average diosgenin content of bulk material during August was 3·86% (Crude) as against 2·11% (Crude) for the material collected in November. Similarly a large number of samples harvested from Jammu plantations in July gave a diosgenin content (pure) above 2·0% (d.w.b.). There appears to be a close relationship between the flowering and fruiting of the plant and diosgenin content.

The foregoing observations indicate a wide range of variability in the diosgenin content of the rhizomes of *Costus speciosus*. There is a definite clonal variation with a very wide range of diosgenin content. The variation appears to be genetic rather than physiologic as no relationship with the size and correspondingly the age of the rhizome could be established. A large percentage of plants growing wild in the sub-Himalayan localities of Jammu, Kangra and Dehradun contain appreciably high content of diosgenin. This makes the task of evolving a high yielding strain more easy.

The abrupt and sharp rise of diosgenin during the months of July and August, an equally sharp decline in September and October and reduction of diosgenin content to less than half of July concentration during the period of dormancy suggests that the crop should be harvested during the peak diosgenin months.

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---

1. Sarin, Y. K., Bedi, K. L. and Atal, C. K., *Curr. Sci.*, 1974, **43** (18), 569.

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LETTERS TO THE EDITOR

HISTAMINE CONTENT OF THE GUT AND VISCERAL ORGANS OF THE FROG *RANA TIGRINA* (DAUD.)

SEVERAL workers have studied the histamine content of the different organs of mammalian species[1-4]. Comparatively little information is available on the histamine content of the lower vertebrates[5]. There are a few reports of the formation and degradation of the histamine and the physiological significance in lower vertebrates[6-7]. In the present investigation histamine content of the gut and certain visceral organs of the frog has been estimated biologically.

Mature frogs of both sexes in a weight range of 100–150 g were decapitated and the tissues were immediately excised and chopped by Mc Ilwain tissue chopper and extracted with 10% trichloroacetic acid for the estimation of the histamine content as per method of Paratt and West[8].

It is evident from Table I that the histamine content in the different portions of the gut of the frog is not consistent. The level of histamine was highest in the stomach and lowest in the ileum. Oesophagus, duodenum and rectum contain histamine in moderate quantities in comparison with the stomach and ileum. Among the visceral organs, the higher levels of histamine was found in lungs followed by kidneys, lever, skin and spleen in the decreasing order.

In the lower vertebrates, the main store of the histamine has been found to be in the tissues and not in the mast cells[9-10]. It has also been suggested that the main function of histamine in the gut of lower vertebrates may be related with the gastric secretory process actively or passively[9]. In the present investigation the histamine content of the stomach has been found to be significantly lower than the reported values for other species of the genus.

The histamine content of the ileum of the frog in the present study is nearer to the values of the histamine content for the other species of the genus *Rana*. However, information about the histamine content of the oesophagus, duodenum and rectum of the other amphibian species has not been reported so far. Histamine content of the lungs of the *Rana tigrina* has been found to be the highest as compared to the previous reports[5]. In the kidney, liver and spleen, a considerable amount of histamine has been detected but its physiological significance in these animals is not clear. In mammals, the kidney and liver are the main sites of inactivation of histamine in the body, being rich in the enzyme histaminase or histamine-N-methyl-transferase. That causes a low level of histamine content in these organs[3]. In the lower vertebrates including certain species of frogs and toads both these enzymes have been reported from liver and kidney[5]. This might be responsible for the low levels of histamine of these organs of the *Rana tigrina* too. Histamine content of the skin in the present study is close to the values reported for the *Rana temporaria,* but it is lower than the values found in the skin of the two species of *Hyla*[5]. The reported values of the histamine content in the other amphibian species are given in Table I for comparison.

TABLE I

Histamine content of the gut and visceral organs of the frog Rana tigrina. *Each figure is mean of five estimations from different animals. Histamine content represented in μg/g of wet weight of tissue ± S.E.*

Sl. No.	Organ	Histamine content	Reported values in other species*
1.	Oesophagus	0·429 ± 0·131	D.N.
2.	Stomach	0·833 ± 0·183	5·5–12·1
3.	Duodenum	0·429 ± 0·122	D.N.
4.	Ileum	0·332 ± 0·072	0·2– 0·3
5.	Rectum	0·454 ± 0·139	D.N.
6.	Lungs	0·433 ± 0·067	0·1– 0·2
7.	Liver	0·348 ± 0·072	D.N.
8.	Kidney	0·356 ± 0·072	D.N.
9.	Spleen	0·171 ± 0·043	D.N.
10.	Skin	0·198 ± 0·042	0·05–0·2

D.N. = Data not reported previously.
* Data from Reite[5].

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1. Haeger, K. and Kahlson, G., *Acta Physiol. Scand.*, 1952, 25, 236.
2. Feldberg. W., In *Histamine* Eds. G. E. W. Wolstenholme and C. M. O.' Conner, Ciba., Found. Symp. Churchil, London 1956. p. 4.
3. Kahlson, G. and Rosengren, E., *Physiol. Rev.*, 1968, 48, 155.
4. Arora, S, Lahiri, P. K. and Kumar, V., *Aspec. Allar. Imun.*, 1968, 2, 105.
5. Reite, O. B., *Physiol. Rev.* 1972, 52, 778.
6. Kimata, M., "The histamine problem", In: *Fish as Food,* Edited by G. Borgstrom, Academic Press, New York, 1961, Vol. I, p. 329.
7. Burkard, W. P., Gey, K. P. and Pletscher, A., *Helv. Physiol. Pharmacol. Acta,* 1961, 19, C 61.
8. Parratt, J. R. and West, G. B., *J. Physiol.* (Lond.), 1957, 137, 169.
9. Reite, O. B., *Experientia,* 1969, 25, 276.
10. Takaya, K., Fujita, T. and Endo, K., *Nature* (Lond.), 1967, 215, 776.

OBSERVATIONS ON HIGH ALTITUDE AND VIVIPARY IN LIZARDS

Agama tuberculata and *Lygosoma himalayanum* are the two very common lizards in Kashmir, which show different altitudinal preferences for their haunts. While *A. tuberculata,* generally does not extend its activity above 6000 ft, *L. himalayanum* stays in pine forests and grassy meadows between 5000–9000 ft (very rarely at 5000 ft). These two distributional belts fall into the Upland Savana and Grassy upland and Alpine meadows respectively—the two life zones of Loveridge (1937), each having its own bio-ecological characteristics.

Loveridge (1937) lists different modes of reproduction in snakes and lizards characteristic of each of his ecological life zone, albeit for East Africa region alone. Subsequent investigations into the reproductive habit of reptiles on latitudinal and altitudinal clines in the Orient and Palearctic region (Sergeev, 1940) not only supplemented the earlier data but concur with broad generalisations of Loveridge (*op. cit.*) that at lower altitudes (his Upland Savana) the preponderance is of oviparous forms which are supplanted by ovoviviparous and viviparous forms in higher reaches (Alpine Meadows etc.).

Agama tuberculata is an oviparous lizard. In exhibiting oviparous habit, it conforms to the reproductive habit characteristic of the family as also to that listed characteristic by Loveridge for the reptilian forms which are distributionally peculiar to Savana Upland.

Geer (1968) and Greene (1970) hold that live bearing habit increases with the altitudinal rise in the habitat of a lizard. That *L. himalayanum,* which is a very common lizard above 6000 to around 9000 ft above sea level, is an ovoviviparous form, is thus neither surprising nor unexpected. The only forms which, however, continue to show oviparous habit even at such high altitudes are a few lacertids and a few geckonids (Geer, 1968), could be, therefore, regarded as exceptional. Although there exists a consistent correlation between vivipary, low temperature and extreme altitudes, Geer (1968), McCoy (1968) and Greene (1970) hold that it is not a relationship of the cause and effect, while Tinkle (1967) hints that vivipary has been an outcome of several other factors that would operate at different altitudes and thereby exercise a selection pressure favouring this mode of reproduction to the other. He does not, however, spell out the various factors involved to which the present authors have to contribute as under.

Our findings show that the total time which *L. himalayanum* spends above ground is barely 6–7 months in a year (May to October), during which period the ambient temperature is of a fairly low order (3°–21° C) in contrast to the temperatures at lower altitudes (8°–35° C). At such high altitudes and at such considerably lowered environmental temperatures an extra-maternal embryonal development as any other bio-chemical process would get slackened and seriously impeded. Added to such a thermal disadvantage, the short duration of barely 6-month of diurnal activity of the lizard at such altitudes would further have cramped the various developmental stages in order that the short span of favourable time in the open available for development of the lizards may not have to pass into the severity of impending winter (October onwards). To obviate such deleterious climatico-ecological effects, the adoption of an intra-maternal development of the embryo has obvious advantage and seems, hence, been selectively favoured. The fact that *L. himalayanum* is only an ovoviviparous form, however, indicates that intra-maternal residence of the developing embryo must originally have been only a protectional adaptation and to that extent may also indicate that it has been a step in the course of evolution of vivipary in lizards. Thus photoperiod as a factor, it appears, has its own role to play in the process. This assumption draws support from similar conclusions from Greene (1968).

While outlining the advantages conferred by vivipary upon the embryo, Greene (*op. cit.*) plausibly argues that the thermo-regulation by basking in cool climates at high altitudes (above 6000 ft) confers an advantage "for embryos to undergo development in the female rather than in an egg abandoned in the environment".

Another equally important factor appears to be the relative fecundity of the concerned forms. *L. himalayanum* has been observed to produce on an average lesser number of all types of eggs (Table I) than *A. tuberculata,* although both show a similar breeding period (May to July) and the latter retains oviducal eggs for two months (from middle of May to middle of July ; no lizard seemed to bear oviducal eggs after July) as against *L. himalayanum,* which retains the oviducal eggs and/or near term embryos for two months and a half (early May to mid July). This works to over 25% longer gestation period in *L. himalayanum.* These facts add the factor of fecundity to the eco-habit differences in the two lizards without, however, reducing the importance of temperature as one of the other factors involved. A longer gestation and reduced fecundity would inevitably render the scincid lizard a comparative disadvantage that would exclude from the company of oviparous species which are potentially better reproducers. With increased tendency towards the accomplishment of vivipary, the ability to produce the greater number of eggs, thus appears to have got progressively reduced ; and with progressive lowering in fecundity, the surest coverage against any risk of ovular dystrophy or failure of fertilized eggs to develop in adverse environmental conditions would be, for the female, to bear the eggs within her body during the embryonal development—a stage identifiable as ovovivipary. Any further intimacy between the embryo and the maternal tissue must ultimately have led to vivipary. Severity of climatic conditions, reduced thermoperiod, low fecundity and high altitude together with other intrinsic factors like hormonal interplay and extrinsic factors like different types of rays that obtain at high altitudes might have collectively contributed to the evolution of vivipary—envisaged as a multifactorial hypothesis as hinted by Tinkle (1967).

TABLE I

Monthly incidence of ovarian and oviducal eggs in Agama and Lygosoma

Month	*Agama tuberculata*		*Lygosoma himalayanum*	
	Ovarian Eggs	Oviducal Eggs	Ovarian Eggs	Oviducal Eggs
April	+/36 (26–40)	..	+*	+/2
May	+/32 (22–34)	+/6	+/16 (12–18)	+/4
June	+/24 (22–26)	+/7	+/18 (12–20)	Emy/6
July	+/22 (18–24)	+/7	+/18 (12–22)	Emy/4
August	+/20 (18–24)	..	+/20 (14–24)	..
September	+/24 (18–26)	..	+/20 (16–24)	
October	+/28 (26–32)	..	+/22 (16–26)	..

* Only one specimen obtained for this month.

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---

1. Greene, H. W., "Mode of reproduction in lizards and snakes of the Gomez Faria region, Tamaulipas, Mexico," *Copeia,* 1970, 3, 566.
2. Geer, A. E., "Mode of reproduction in the squamata fauna of three altitudinally correlated life-zones in East Africa," *Herpetologica,* 1968, 24, 229.
3. Loveridge, A., "Scientific results of an expedition to rain forest regions in Eastern Africa. IX, Zoo-geography and itinerary," *Bull. Mus. Comp. Zool.,* 1937, 79 (9), 481.
4. McCoy, C. J., "Reproductive cycle and viviparity in Guatemalan, *Corythophanes percarinatus,*" *Herpetologica,* 1968, 24, 175.
5. Sergeev, A. M., *Researches on Viviparity of Reptiles 135th Anniv. Pub. Moscow, Soc. Nat.,* 1940.
6. Tinkle, D. W., "The life and demography of the side-blotched lizard *Uta stansburiana stejnegeri,*" *Misc. Publ. Mus. Zool. Univ. Michigan,* 1967, 132, 1.

PHYSIOLOGICAL STUDIES ON THE EFFECTS OF NUTRITIONAL IMBALANCE ON THE CENTRAL NERVOUS SYSTEM: HORMONE-LIKE ACTION OF THE BLOOD OF THIAMINE-DEFICIENT CHICKEN, *GALLUS DOMESTICUS*

IT is well established that the deficiency of B_1-vitamin (Thiamine) causes beri-beri in man and signs of nervous symptoms in birds referred to as Poly-neuritis[1-2]. Thiamine as thiamine-pyrophosphate is known to influence the oxidative decarbolation of pyruvate in pigeon and man[3,4]. It has been suggested that accumulation of pyrurate in thiamine-deficient vertebrates may be responsible for various signs of poly-neuritis[4,5]. It is presumed that injury to medulla is responsible for the pathogenic symptoms of B_1-avitaminosis. The medullary nervous symptoms during thiamine-deficiency have been reported to be due to toxic product accumulation[5,6].

Earlier investigations of Nayeemunnisa[7,8] and other workers[9] indicated changes in the levels of RNA, activity levels of certain oxidative enzymes and acetylcholinesterase in the different regions of the brain of chicken, *Gallus domesticus,* during thiamine-deficiency. The present investigation aims to see whether the neural disturbances result in blood-borne factors and if so whether these factors affect the cardiac rhythmicity.

Three days old, male white leghorn chicken, *Gallus domesticus,* ranging in weight from 12–14 grams were reared in the laboratory in electrically-heated cages at 37 ± 2° C. The controls were fed on standard chicken feed (Mysore Feeds, Bangalore, India). The experimental birds were fed on polished rice for 25–30 days as described by Peters[5] to induce thiamine-deficiency. Water was made available *ad libitum* to both the groups.

The control chicken were given mild ether anaesthesia and dissected with sterilized instruments to expose the heart. The heart and the exposed body cavity was flooded with avian Ringer[10,11] at the body temperature. The heart was then carefully connected with the help of a wire hook and thread to the writing pointer of the kymographic drum (Patent Inco, India. No. M.E. 1880 220 Volts A.C. rotated at 2·5 mm/second) after *in vitro* soaking by blood obtained from other control chicken. These recordings of the cardiac contractions constituted observations for the control animals. The preparation was washed in fresh avian Ringer[11]. Blood from the thiamine-deficient chicken, showing signs of opisthotonus was extracted from the wing vein with a sterilized syringe, was added directly on the heart of the control chicken and the heart beat under the influence of the blood of avitaminous birds was recorded kymographically under identical conditions.

The kymographic recordings were fixed in turpentine : varnish (2 : 1) and photographed. In general the heart beat is decreased on *in vitro* administration of blood from thiamine deficient chicken. The per cent decrease of heart beat in control birds is – 26·4, – 50·0 and – 28·2 in the burst frequency (33 and 24·3 per minute for controls and experimentals respectively) amplitude (1·5 and 0·75 mm for controls and test birds respectively) and duration (2·2 and 1·6 seconds for controls and experimentals respectively) whereas inter-burst interval was prolonged (per cent increase is 92·3 and average values being 0·13 and 0·2 mm for controls and test animals respectively) on administration of blood from B_1-avitaminosis birds (Table I).

TABLE I

The heart-beat parameters of Gallus domesticus

Characteristics measured	*Controls	**Experimentals	Percentage change
Burst frequency (Per minute)	33·0±3·0	24·3±2·7	– 26·4
Burst amplitude (mm)	1·5±0·21	0·75±0·23	– 50·0
Burst duration (Seconds)	2·2±0·01	1·58±0·016	– 28·2
Inster-burst Interval (mm)	0·13±0·02	0·25±0·01	+92·3

Values are Mean of 4 observations ± S.D.

(+) and (–) signs indicating increase and decrease respectively over controls.

Values are significant at $P > 0\cdot01$.

* Control chicken heart soaked with the blood of normally active chicken.

** Control chicken heart soaked with the blood of thiamine deficient chicken.

These changes produced in the heart beat by the blood of thiamine deficient chicken, *Gallus domesticus* clearly indicate the presence of one or more hormone-like factors which can produce *in vitro* effects. Generally, thiamine deficiency results in the profound changes in metabolic rate of the tissues[3-5]. The fact that the blood of avitaminosis birds is capable of depressing the heart beat of normal chicken, clearly suggests the presence of an inhibiting factor in the blood of thiamine-deficient birds, produced as a consequence of metabolic alterations.

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1. Eijkman, C., *Genisk Tijkschrifit Ned. Ind.*, 1890, 30, 295.
2. Grijns, G., *Geneesk. Tijdscht. Ned. Ind.*, 1901, 43, 3.
3. Harper, H. A., *Review of Physiological Chemistry*, Maruzer Asian, Edition, 1965.
4. Fruton, J. S. and Simmonds, S., *General Biochemistry,* Asian Student Edition, Asia Publishing House, Madras, 1960.
5. Peters, R. A., *Biochemical Lesions and Lethal Synthesis,* International Series of Monographs on Pure and Applied Biology, Pergamon, Student, Edition, 1963, 18.
6. Walshe, F. M. R., *Med. Sci. Abstr.*, 1920, 2, 411.
7. Nayeemunnisa, *Life Sciences,* Part III, Communicated, 1975.
8. —, *Experientia,* Communicated, 1975.
9. Nalini, U. P. and Nayeemunnisa, *Ibid.*, 1976, 32 (2), 198.
10. Prosser, C. L. and Brown, F. A., *Comparative Animal Physiology,* Saunders Company, Philadelphia, 1961.
11. Cavanaugh, G. M., *Formulae and Methods,* Marine Biological Laboratory, Woods Hole, Mass., 1956.

PEROXIDASE ACTIVITY IN PAPAYA PLANT INFECTED WITH PAPAYA DISTORTION MOSAIC VIRUS

VIRAL infection induces several metabolic changes in plants[1,2]. Changes in peroxidase activity following infection have also been reported. Loebenstein and Linsey[3], Bhargava *et al.*[4], and Wood and Barbara[5] reported an increase in peroxidase activity in vein clearing virus infected sweet potato, sugarcane plants infected with sugarcane mosaic virus and tobacco plants infected with cucumber mosaic virus respectively. In tobacco mosaic virus infected tobacco leaves, however, peroxidase activity decreased below that of healthy leaves both during early and later stages of the development of the disease[6]. Present investigation was undertaken to examine the effect of papaya distortion mosaic virus on peroxidase activity in papaya leaf.

To determine the effect of papaya distortion mosaic virus on peroxidase activity, papaya plants grown in the glass house were inoculated at 4–5 leaf stage with sap from infected papaya plants using carborandum as an abrasive. Healthy plants of the same age served as control. Peroxidase activity was determined at 0, 3, 6, 9, 12 and 15 days after inoculation. During sampling 6 discs of 1 cm diameter each were taken from healthy as well as infected leaves, care being taken that sampling was always done from the third leaf. The discs were weighed and homogenized in 2·0 ml of chilled distilled water and filtered through cheese cloth. The filtrate was centrifuged in a refrigerated centrifuge at 1,000 rpm for one hour. The enzyme activity in the clear supernatant was measured colorimetrically according to the method of Loebenstein and Linsey[3] as described by Wood and Barbara[5].

The results presented in Table I show that the peroxidase activity in the papaya leaves increased with papaya distortion mosaic virus infection, the increase being upto a maximum of 39·5% after 9 days of inoculation.

TABLE I

Peroxidase activity in healthy and virus infected papaya leaves

Days after inoculation	Enzyme activity (S^{-1} g^{-1} fresh wt.)		
	Control	Inoculated	Percentage increase
0	27·1	27·1	0
3	21·3	26·7	25·3
6	22·0	25·7	16·9
9	13·4	18·7	39·5
12	23·2	31·3	34·9
15	38·9	37·5	0

Peroxidase is known to catalyse the oxidation of phenolic substances and aromatic amines to quinones in the presence of hydrogen peroxide[7], though its exact physiological role, even in normal metabolism, is not clearly understood. In many cases increase in peroxidase activity in plants has been correlated with either disease resistance[8,9] or susceptibility and symptom response to virus[3,10]. Loebenstein and Linsey[3] reported that increase in peroxidase activity following inoculation commences with the appearance of symptoms in the infected plants. In our study, increase in peroxidase activity was found after 3 days, when no symptoms had appeared though viral activity was present. Maximum increase was. however, found after 9 days of inoculation. In this connection, it may be mentioned that viral activity was also more pronounced between 9 and 12 days after infection, showing a correlation between viral activity and increase in peroxidase activity due to infection.

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1. Bawden, F. C., *Ann. Rev. Plant Physiol.*. 1959, 10, 239.
2. Diener, T. O., *Ann. Rev. Phytopath.*, 1963, 1, 1970.
3. Loebenstein, G. and Linsey, N., *Phytopathology*, 1961, 51, 533.
4. Bhargava, K. S., Joshi, R. D. and Srivastava, G. P., *Experientia*, 1970, 26, 216.
5. Wood, K. R. and Barbara, D. J., *Physiology Plant Pathology*, 1971, 1, 73.
6. Wynd, F. L., *Jour. Gen. Physiol.*, 1942, 25, 649.
7. Bonner, J., *Plant Biochemistry*, Academic Press, N.Y., 1950, p. 537.
8. Fehrmann, H. and Dimond, A. E., *Phytopathology*, 1967, 57, 69.
9. Simons, T. J. and Ross, A. F., *Ibid.*, 1970, 60, 383.
10. Menke, G. H. and Walker, J. C., *Ibid.*, 1963, 53, 1349.

INHIBITION OF THREE STRAINS OF WATERMELON MOSAIC VIRUS BY BARK EXTRACTS*

SEVERAL plant virus inhibitors have been reported to be present in different parts of the higher plants (Dugger and Armstrong[3], Johanson[6], Gendron[5], Sill and Walker[9]. Bawden[1], Crowley[2]. Feldman[4], Ulrichova and Bracak[11]). Little attention has been given to the inhibitory activity of juices from plant barks. Recently Singh[7,8] studied the effect of several bark extracts on the inactivation of Tobacco mosaic virus (TMV) and poptato virus X, and has shown the presence of inhibitors in the aqueous extracts of the barks. Tamura[10] reported the inhibitory activity of bark extracts of Japanese black pine—*Pinus thunbergii* on the infectivity of turnip mosaic virus.

The present study deals with the effect of the bark juices of nine trees belonging to different families on the infectivity of WMMVB, WMMM and WMMC strains of watermelon mosaic virus.

Standard inoculum of virus strains used in the present study were prepared by macerating young infected leaves of *Cucurbita pepo* L. cv. Caserta showing good symptoms. The crude juice was squeezed through layers of muslin cloth. Bark from different plants was collected separately. In each case 20 g of clean, fresh bark in the form of small pieces were homogenized with an equal volume of distilled water in a Waring Blender. Juices from these homogenates were expressed through a muslin cloth and were mixed with standard inoculum in 1 : 1 ratio. This mixture was then rubbed on healthy leaves of *Cucurbita pepo* L. cv. Caserta. Standard inoculum mixed with distilled water in 1 : 1 ratio was used as control. Five replicates were taken for each treatment. The results of inhibition of three strains of watermelon mosaic virus by bark extracts are given in Table I.

TABLE I

Effect of bark extracts of trees on the infectivity of three strain of watermelon mosaic virus

Species	Virus Strains	Number of plants infected out of 50 inoculated	Percentage of inhibition
Azadirachta indica L.	WMMVB	30	40
	WMMM	40	20
	WMMC	40	20
Callistemon lanceolatus D.C.	WMMVB	45	10
	WMMM	40	20
	WMMC	50	0
Eriobotrya japonica Lindle.	WMMVB	40	20
	WMMM	40	20
	WMMC	45	10
Ficus bengalensis L.	WMMVB	20	60
	WMMM	20	60
	WMMC	20	60
Ficus elastica Roxb.	WMMVB	10	80
	WMMM	10	80
	WMMC	20	60
Grewia asiatica L.	WMMVB	0	100
	WMMM	0	100
	WMMC	0	100
Prunus communis L.	WMMVB	40	20
	WMMM	40	20
	WMMC	40	20
Syzigium cumini D.C.	WMMVB	10	80
	WMMM	20	60
	WMMC	0	100
Tamarindus indica L.	WMMVB	10	80
	WMMM	20	60
	WMMC	10	80

Results in Table I indicate that bark extract of *Grewia asiatica* L. is most inhibitory to the infectivity of all three strains of watermelon mosaic virus. It is followed by extracts of *Syzigium cumini* D.C., *Tamarindus indica, Ficus etastica* Roxb., *Ficus bengalensis* L., reduced the infectivity by more than fifty per cent. *Azadirachta indida* L., *Prunus communis* L., *Eriobotrya japonica* Lindle, and *Callistemon lanceolatus* D.C., showed mild inhibitory activity.

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1. Bawden, F. C., *Adv. Virus. Res.*, 1954, 2, 31.
2. Crowley, N. C., *Aust. J. Bot.*, 1955, 8, 56.
3. Dugger, B. M., and Armstrong, J. K.. *Ann. Mo. Bot. Gdn.*. 1925, 12, 359.
4. Feldman, J. M.,** *Ann. Epiphyt.*, 1963, 14, 377.
5. Gendron, Y.,** *Acad. Sci. Paris,* 1960, 230, 1974.
6. Johanson, J., *Phytopathology,* 1941, 31, 676.
7. Singh, R., *Experientia,* 1969, 25, 218.
8. —, *Phytopathologia medit,* 1971, 10, 2111.
9. Sill, W. H. and Waler, J. C., *Phytopathology,* 1952, 42, 349.
10. Tamura, M., *Ann. Phytopath. Soc,* Japan, 1969, 36, 260.
11. Ulrychova, M. and Bracak, J., *Phytopath. Z.,* 1967, 58, 87.

**Original not seen.

PACHYTENE CHROMOSOMES OF JUTE (*CORCHORUS CAPSULARIS*)

THE pachytene chromosomes of *Corchorus olitorius* were studied by Joshua, Thakare and Rao[1] and Paria and Basak[2]. These studies were found to agree, in general, on the morphology of the chromosome complement. The present study deals with the pachytene chromosomes of *C. capsularis* and attempts to find out whether the incompatibility between these species is associated with gross chromosomal differences.

Flower buds of *C. capsularis* var. JRC 412 were fixed in 1 : 3 acetic alcohol and slides were prepared following the methods employed for *C. olitorius*[1]. Microphotographs were taken at × 1,000 and the negatives were enlarged four times. The data on each chromosome are based on the measurements of 5 to 7 well-spread preparations.

The pachytene chromosomes of *C. capsularis,* although significantly longer (Table I) than those of *C. olitorius*[1], resemble those of the latter in many respects (Figs. 1, 2). The relatively shorter chromatic segments composed of conspicuous, deeply staining chromomeres flanking the centromeres and the long, achromatic distal segments are characteristic of both *C. olitorius*[1] and *C. capsularis.* All the chromosomes except chromosome 2 (nucleolar) and chromosome 3 have a median centromere. The nucleolar chromosome is the only one with a submedian centromere in both the species. The pattern of distribution of the chromomeres also seem to be similar, with *C. capsularis* having a few additional chromomeres. The average arm length ratio of the complement is 1·74 in *C. olitorius and* 2·10 in *C. capsularis.* The achromatic to chromatic ratio for the individual chromosomes is comparable to those of *C. olitorius*; the average ratio for the complement is 2·08 and 2·09 for *C. olitorius* and *C. capsularis* respectively. The ratio between the longest and the shortest chromosome of the complement in *C. capsularis* is 1·66 which is not significantly different from 1·55 of *C. olitorius.*

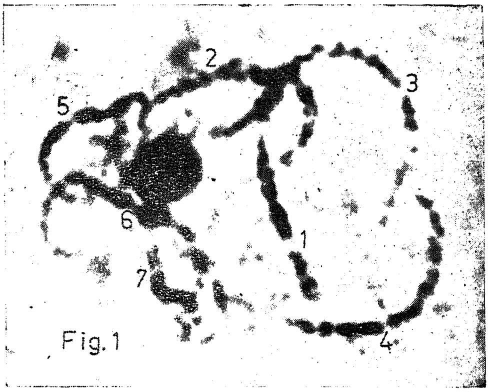

FIG. 1. Pachytene complement of *Corchorus capsularis.*

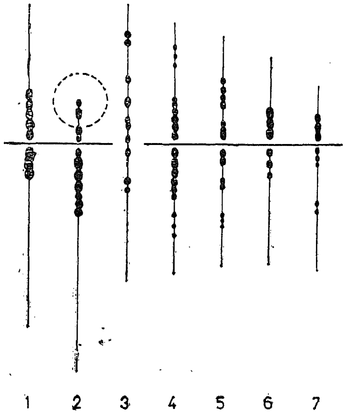

FIG. 2. Idiogram of pachytene chromosomes of *Corchorus capsularis.*

Despite these striking similarities between the pachytene complements of the two species, minor differences are observed when the chromosomes are studied individually. Some of the chromomeres in the achromatic regions are more prominent in

TABLE I

Total length, arm ratio and achromatic to chromatic ratio of pachytene chromosomes of Jute (each value is mean of 5–7 cells)

Chromosome No.	Total length (μ)	Length of short arm (μ)	Length of long arm (μ)	Arm length ratio	Length of chromatic segment (μ)	Length of achromatic segment (μ)	Achromatic to chromatic ratio
1.	26·88	11·65	15·23	1·31	7·23	19·65	2·72
2.	22·56	3·70	18·86	5·10	9·64	14·19	1·07
3	23·27	..	..	..	5·80	17·47	3·01
4.	20·63	9·94	10·69	1·08	10·29	10·34	1·00
5.	20·00	8·90	11·10	1·25	5·79	14·21	2·45
6.	17·29	7·12	10·17	1·43	5·00	12·29	2·46
7.	16·17	5·34	10·83	2·03	5·47	10·70	1·96

C. capsularis. An increase in the length of the achromatic segments is also observed in this species. These differences, however, appear to be genotypic and do not seem to account for the strong cross incompatibility between the two species.

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---

1. Joshua, D. C., Thakare, R. G. and Rao, N. S., *Indian J. Genet.*, 1973, 33, 452.
2. Paria, P. and Basak, S. L., *Curr. Sci.*, 1973, 42 (23), 832.

ON THE FORMATION OF THE MICROPYLE IN *NEMACHILUS TRIANGULARIS* DAY

DURING a detailed investigation on the anatomy and histology of the reproductive system of the common loach *Nemachilus triangularis* Day it was possible to observe the origin and formation of the micropyle. At the migratory nucleus stage the oocyte is spherical and measures 500 μ in diameter. The germinal vesicle which is very near the animal pole appears as a very prominent oval body with a distinct wall which is thrown into feeble folds (Fig. I). A distinct zona radiata covers the oocyte all around. This egg membrane has a large number of closely packed striations giving it a tough consistency. Over this, the follicular epithelium forms a single layered membrane, the cells being cuboidal. Each cell has a very distinct central nucleus with dense cytoplasm Immediately dorsal to the germinal vesicle, the micropyle is visible as a funnel-shaped depression of the zona radiata, with the wide end directed outwards. Three cells of the follicular epithelium lying dorsal to the germinal vesicle get enlarged and appear differently (Fig. 2). The nucleus in each of these three cells is eccentric in position. The portion of the cell dorsal to the nucleus gets vacuolated and the cell walls between them disappear. The cytoplasm of

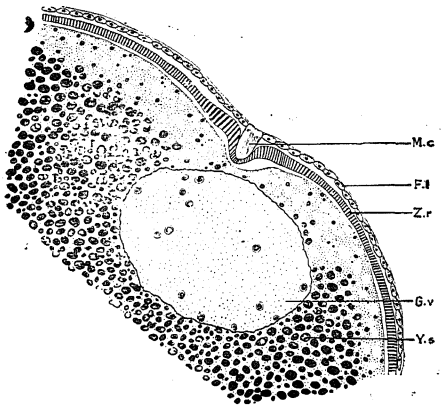

FIG. 1. T.S. of migratory nucleus stage of oocyte of *Nemachilus triangularis* showing strands of protoplasm in the micropylar cell. (Reconstructed from 10 consecutive serial sections) × 250.

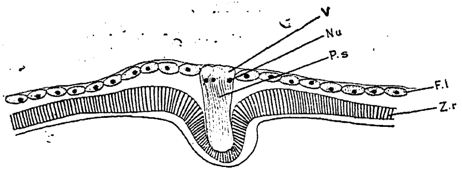

FIG. 2. T.S. of oocyte passing through the micropylar region showing follicular layer. micropylar funnel, and zona radiate, × 400.

all the three cells are thrown into fibrillar strands which run parallel to one another from the base of the nucleus to the posterior end of the coalesced cells. In section, the whole structure appears as a rectangular stringed plug, the posterior part of which firmly pressing the zona radiata downwards. This inpushing results in the formation of a deep depression, the micropyle.

The formation of the micropyle in teleosts has been studied by a few earlier workers[1-5] who have come to the conclusion that the micropyle owes its origin to the follicular cells. In *Pygosteus pungitius*[2] and in *Macropodus cupanus*[5] a single follicular cell gets modified and the posterior part of this cell is drawn into a wedge-shaped process which presses down the zona radiata to form the micropyle. In *Leucius*[3] three follicular cells crowd together and press the zona radiata into a funnel-shaped structure forming the micropyle. In *Stigmatogobius javanicus*[1] a cluster of cells of the follicular layer exerts pressure in the zona radiata to form the micropyle. In the present study however it can be observed that the micropyle is formed by the coalescence of three enlarged follicular cells, the cytoplasm of which is thrown into firm strands, that coalesce posteriorly.

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1. Aravindan, C. M. and Padmanabhan, K. G., *Acta Zoologica*, 1972, 53, 45.
2. Eigenmann, C., *Bull. Mus. Harv. Col.*, 1890, 19 (2), 129.
3. Hoffmann, C. K., *Verhandl. d. koninkl. Akad. V. Wetenschappen.* Amsterdam., 1881, Deel XXI. 164 pp, 7 Taf.
4. Mark, E. L., *Bull. Mus. Harv. Col.*, 1890, 19(1).
5. Padmanabhan, K. G., *Bull. C.R.I.*, Trivandrum, 1955, 4 (1). 1.

CHARACTERIZATION OF NUCLEAR HISTONES IN SPERMATOZOA OF A CENTIPEDE (*MECISTOCEPHALUS* SP.)

Introduction

THERE are conflicting reports in literature regarding dual nature of the testes and spermatogenesis in Chilopods, leading to the formation of two types of Spermatozoa. Bouin (1925) reported two types of spermatogenic processes in the same testis of *Scolopendra cingulata.* Dual nature of testis having two types of spermatozoa have also been reported by Ansley (1954) in *Scutigera forceps.* However Nath (1925), Ram (1937) and Gulati (1937) in their contribution on the spermatogenesis of *Lithobius forficatus, Rhysida longipes* and *Scolopendra* sp., respectively, do not reveal such duality of spermatogenesis in anyone of the three forms.

In *Mecistocephalus* sp., under light microscopic work, no morphological differences are seen in sperm-cells, but there may be differences at the chemical level and also in their viability and fertilizing ability.

In the present investigation, cytochemical analysis of the nuclear histones has been carried out with the idea to ascertain if two types of spermatozoa are formed. It may then be possible to distinguish them according to the type of basic nuclear proteins present in their head-part. It is now well established that during spermatogenesis, a typical somatic or lysine rich histone is replaced by arginine rich histone type, at spermatid stage and in some species there is further alteration to protamine in mature spermatozoa (Dupraw, 1970). The role of basic nuclear protein in the sperm-head still remains unknown (Bloch, 1969).

Materials and Methods

Specimens of *Mecistocephalus* sp. were collected from damp soils of the Western Himalayan region. The testes and seminal vesicles from the male centipedes and oviducts from the female were dissected out in physiological saline solution.

Spermatozoa were taken out from these organs by teasing apart the tissues in physiological saline and were spread out in the centre of a few properly cleaned slides. The smears thus prepared were air-dried and slides were processed in the procedure stated below.

The reagents used to stain the basic nuclear protein of sperm-cells consisted mainly of trichloroacetic acid (TCA)- alkaline fast green method as prescribed by Alfret and Geschwind (1953), and picric acid-bromophenol blue method of Bloch and Hew (1960). Such procedures can be used to differentiate among somatic histones, arginine rich and protamines.

The Alfret and Geschwind method is used specifically for the detection of nuclear histones at a pH 8·0–8·02, because any protamines present in the cell would be extracted by hot TCA. However, protamines are stained by bromophenol blue (at pH 2·3) after treatment with picric acid. Lysine rich and arginine rich histones stain with alkaline fast green or bromophenol blue. The lysine rich histones lose most or all their stainability after deamination (Van slyke, 1971) or acetylation

TABLE I

Characterization of nuclear histones of spermatozoa

Method	Spermatozoa from the testes	Spermatozoa from the seminal-vesicles	Spermatozoa from the oviducts	Conclusion
TCA-alkaline fast green	+	+	+	Protamine absent, lysine or arginine rich histones present
TCA-alkaline fast green after acetylation or deamination	+	+	+	Arginine rich histones
Bromophenol-blue picric acid	+	+	+	Arginine or Lysine rich histones
Bromophenol-blue picric acid after acetylation or deamination	+	+	+	Arginine rich histones

Symbol used = + = Positive reaction.

(Monne and Slautterback, 1950), but the arginine rich histones remain uneffected.

Results and Discussion

Results of the cytochemical studies on nuclear histones of spermatozoa are summarized in Table I.

All the sperm-cells taken out from the testes reacted positively to TCA-alkaline fast green test, indicating that protamines are absent and lysine or arginine rich histones are present. Similar results were observed in all the sperm-cells teased apart from the seminal vesicles of male and oviduct of the female. Staining with Bromophenol blue also gave a positive test. Even after deamination or acetylation the sperm-cells from different reproductive organs reacted positively.

Thus spermatozoa of *Mecistocephalus* sp. are rich in arginine and there is no subsequent shift to protamines. None of the spermatozoa examined from different male and female reproductive parts revealed any contrast in staining.

Hence, the nature of basic nuclear protein does not indicate two different types of spermatozoa in *Mecistocephalus* sp.

However it is remarkable to note that in mature spermatozoa there is a transition from somatic or lysine rich to argine rich histones, but there is no further alteration to protamines. Similar results were obtained in *Drosophila* sperm-cells by Das *et al.* (1969). It will be interesting to find the stage at which the transition occurs from lysine rich to arginine rich during the spermatogenesis.

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1. Alfret, M. and Geschwind, I. I., *Proc. Natl. Acad. Sci.*, U.S.A., 1953, 39, 991.
2. Ansley, H. R., *Chromosoma*, 1954, 6, 656.
3. Bloch, D. P., *Genetics Supplement*, 1969, 61, 93.
4. —, and Hew, H. V. C., *Biophys. Biochem. Cytol.*, 1960, 8, 69.
5. Das, C. C., Gay, H. and Kaufman, B. P., *Exp. Cell Res.*, 1964, 35, 507.
6. Dupraw, E. J., *DNA and Chromosomes*, Holt, Rinehart and Winston, Inc, N.Y., 1970.
7. Gulati, R. K., *J. Roy. Micro. Soc.*, 1937, 57, 237.
8. Monne, L., and Slautterback, D. B., *Ark. Zool. Ser.*, 1950, 1 (3), 455.
9. Nath, V., *Proc. Camb. Philos. Soc.*, 1925, 1, 270.
10. Ram, G., *J. Roy. Micro. Soc.*, 1937, 57, 237.
11. Van, slyke, D. D., *J. Biol. Chem.*, 1911, 9, 185.

ACTIVATION OF PROPHENOLOXIDASE IN THE HEMOLYMPH OF *CALLIPHORA VICINA* R–D

PHENOLOXIDASE (E.C. 1·10·3·1 : *o*-Diphenol : Oxygen oxido-reductase) is present in the hemolymph of *Calliphora vicina* R–D in an inactive form, the prophenoloxidase[1,2]. It is activated by a cuticular activator present in the 3rd instar larva[3,4] or by an endogenous activator present in the hemolymph[5-7]. However, sometimes activation takes place spontaneously, in some insects at certain age[8-10], or by treatment with proteolytic enzymes[2,11,12], metal ions[9,13], alcohols and acetone[12,13]. Therefore, experiments were carried out to find out whether the prophenoloxidase of *C. vicina* also undergoes induced activation, by such substances.

Preparation of prophenoloxidase, cuticular activator (for control) and determination of phenoloxidase activity were done according to standard methods[4], except for some modifications. The pellet obtained after 144,000 g centrifugation was dissolved in 5 ml phosphate buffer (0·05 M, pH 6·5) and 100 microlitre of it were mixed with 4 ml of Dopa (2·4-Dihydroxyphenylalanin 0·5 mg/ml of phosphate buffer) and 100 microlitre of cuticular activator (control) or the testing substance (acetone, alcohol, proteolytic enzyme, metal ion salts 0·01 M). The color development was measured in terms of O.D. using mercury filter of 492 mμ and 20 mm cells in Eppendorf spectrophotometer, while the rate of dopa oxidation was measured at 35° C for 10 minutes and was recorded by an automatic recorder. Enzyme blanks were also run and found non-reactive, during preliminary experiments.

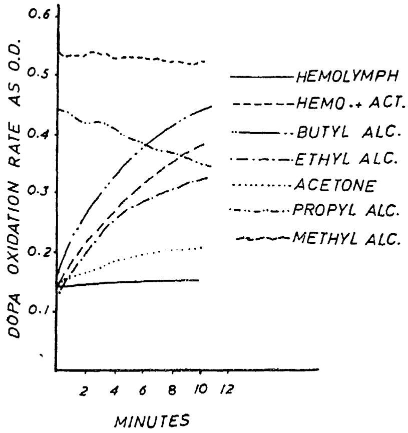

FIG. 1. Effect of alcohols and acetone on the activation of prophenoloxidase in comparison with and without activator.

Experiments showed (Fig. 1) that acetone produced slight activation while ethyl alcohol and butyl alcohol produced significant activation. Methyl and propyl alcohols had no effect. Such an activation has been reported for other insects[12,13]. However, activation by ethyl and butyl alcohols only and not by methyl and propyl alcohols is strange. It may be due to some structural affinity.

Figure 2 indicates that trypsin activated the prophenoloxidase while pepsin did not. Activation by alpha-chymotrypsin and dipeptidase has been reported in *Calliphora*[2], while by alpha-chymotrypsin in the case of *Bombyx mori*[11]. Although pepsin and trypsin both are proteolytic, the former had negligible effect. It may be due to the difference in the optimum pH. This also indicates that there exists proteolytic mechanism for prophenoloxidase activation but under certain optimum conditions.

Activation of prophenoloxidase by metal ions has been reported by a few workers[9,13]. The role of cupric ion has been discussed in the case of silkmoth[14]. During the present investigations it was observed that NaCl, $MgCl_2$, $MnCl_2$, and $MnSO_4$ did not activate the prophenoloxidase (Fig. 3), although Mg^{++} and Mn^{++} increase the activity of many enzymes. However, $FeCl_3$, $CuCl_2$, $FeSO_4$ and $CuSO_4$ activated the prophenoloxidase to a great

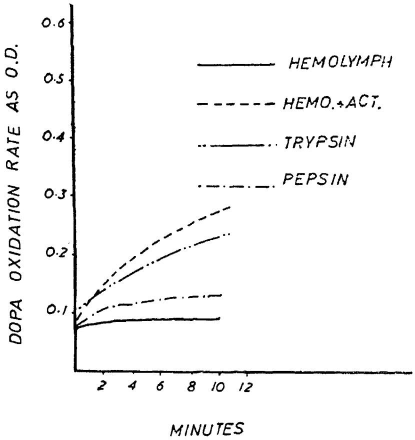

FIG. 2. Effect of pepsin and trypsin on the activation of prophenoloxidase in comparison with hemolymph only and hemolymph + activator.

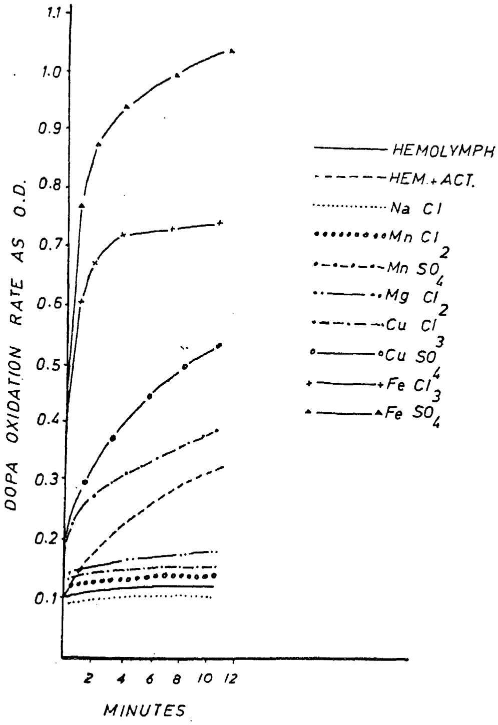

FIG. 3. Effect of different salts of metal ions on the activation of prophenoloxidase in comparison with hemolymph extract (prophenoloxidase) and the hemolymph + activator.

extent. It confirms the findings of Ashida[14] and Naqvi and Ahmad[9]. Ashida[14] has discussed in detail, the probable role of cupric and cuprous ions in the activation mechanism while Naqvi and Ahmad[9] histochemically showed the presence of a cupric or ferric ion dependent isozyme of diphenoloxidase, in polyacrylamide gels. Moreover, the presence of copper in the insect blood and the decrease in the diphenoloxidase activity (one-third) due to conversion of cupric form to cuprous in the silkworm hemolymph[14], on standing, indicates some role of these ions in the prophenoloxidase activation mechanism, at least for one isozyme.

The author is grateful to Prof. P. Karlson, for providing the facilities at the Institute of Physiologische-Chemie, Marburg and Alexander von Humboldt-Stiftung, West Germany, for post-doctorate fellowship. He is also thankful to Dr. M. H. Kasim, Chairman, Biology Department, of this college, for some help.

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1. Karlson, P. and Schweiger, A., *Hoppe–Seyler's Z. physiol. Chem.*, 1961, 323, 199.
2. Schweiger, A. and Karlson, P., *Ibid.*, 1962, 329, 210.
3. Sekeris, C. E. and Mergenhagem, D., *Science*, 1964, 145, 68.
4. Karlson, P., Mergenhagen, D. and Sekeris, C. E., *Hoppe–Seyler's Z. Physiol. Chem.*, 1964, 338, 42.
5. Ohnishi, E., *Annot. Zool. Jap.*, 1954, 27, 33.
6. —, *J. Insect Physiol.*, 1959, 3, 219.
7. Mitchell, H. K. and Weber, A. M., *Science*, 1965, 148, 964.
8. Thomson, J. A. and Sin, Y. T., *J. Insect Physiol.*, 1970 16, 2063.
9. Naqvi, S. N. H. and Ahmad, I., *Baghdad Bull. Sci.*, 1975, 7, (In press).
10. —, Karlson, P. and Novak, H., *Pak. J. Scient. Ind. Res.* 1975 (In press).
11. Ohnishi, E., Dhoko, K. and Ashida, H., *Arch. Biochem. Biophys.*, 1970, 139, 143.
12. Preston, J. W. and Taylor, R. L., *J. Insect Physiol.*, 1970, 16, 1729.
13. Evans, J. J. T., *Ibid.*, 1967, 13, 1699.
14. Ashida, M., *Arch. Biochem. Biophys.*, 1971, 144, 749.

A SPONTANEOUS ALLOTETRAPLOID *ALOE*

The genus *Aloe* L. forms the largest group amongst the three major genera of the succulent plants belonging to the tribe Aloineae. In India, aloes are mostly cultivated as 'vegetative ornamentals' being easily propagated through suckers. While studying the chromosomes of the tribe Aloineae, Brandham[1] has recently analysed 113 species of *Aloe*, and recorded 106 diploids ($2n = 2x = 14$), one triploid ($2n = 3x = 21$), five tetraploids ($2n = 4x = 28$) and a hexaploid ($2n = 6x = 42$). In addition, there are some previous reports of polyploidy in *Aloe*, and Resende[2] has recorded a pentaploid ($2n = 5x = 35$) forma *gigas* of *A. ciliaris* known only in cultivation. Sharma and Mallick[3] have recorded a triploid of *A. humilis*, while Snoad[4] has recorded three hexaploid species of *Aloe*. Thus, the total known polyploids among 113 species of *Aloe* is 12, *i.e.*, the genus approximately shows 10·6% incidence of polyploidy.

The present plant is a new addition to the list of polyploids in *Aloe* and is perhaps the first case of spontaneous allotetraploidy in the Aloineae. This plant shows *gigas* features in some of the morphological characters. It is unique in having a globose condensed raceme type of inflorescence with greenish-yellow flowers, while all the tetraploids known so far have long racemes with red flowers (Brandham, personal communication). Root tip squashes made using Brandham's technique[5] clearly show $2n = 4x = 28$, comprising 16 long and 12 short chromosomes (Fig. 1). Following the classification of the long chromosome pairs in the Aloineae by Snoad[4], the $4\,L_1$ chromosomes appear to be of two different lengths (Fig. 1, hexagons and squares represent two sets of homologues), indicating that the chromosome complement comprises two different genomes. The plant thus appears to be an allotetraploid and was probably a hybrid at the

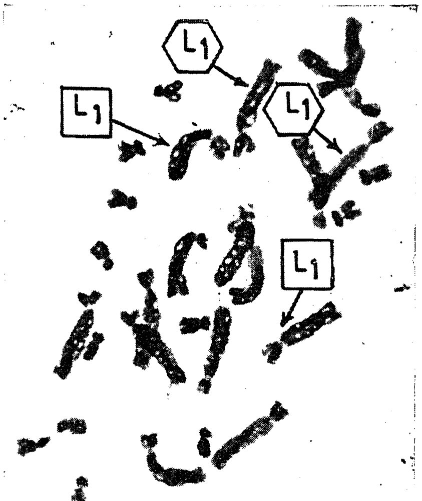

FIG.-1

Fig. 1. Photomicrograph of somatic metaphase showing $2n = 4x = 28 = 16\,L + 12\,S$. Hexagons and squares show homologous L_1 'chromosome sets in the complement. Magnification, × 1,740.

diploid level. Although with allotetraploidy fertility is generally restored[6], this plant is completely sterile and thus presents a case of genic sterility[7]. The diploid hybrid has been studied cytologically[8] and a cytological account of the allotetraploid has been completed.

I am grateful to Prof. K. B. Deshpande. for facilities and to Dr. P. E. Brandham, Jodrell Laboratory, Kew, for advice.

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1. Brandham, P. E., *Kew Bulletin*, 1971, 25 (3), 381.
2. Resende, F., *Ber. Deutsch. Bot. Ges.*, 1938, 56, 533.
3. Sharma, A. K. and Mallick. R., *Jour. Genet.*, 1965, 59, 110.
4. Snoad, B., *Heredity*, 1951, 5, 279.
5. Brandham, P. E., *Chromosoma*, 1975, 51, 269.
6. Burnham, C. R., *Discussions in Cytogenetics*, Burgess Publishing Co., Minnesota 1962.
7. Swanson, C. P., *Cytology and Cytogenetics*, Macmillan and Co. Ltd., Bombay, 1957.
8. Spare, A. B., *Cytologia*, 1975, 40, 797.

HYBRIDIZATION BETWEEN *AMARANTHUS GRAECIZANS* AND *A. VIRIDIS*

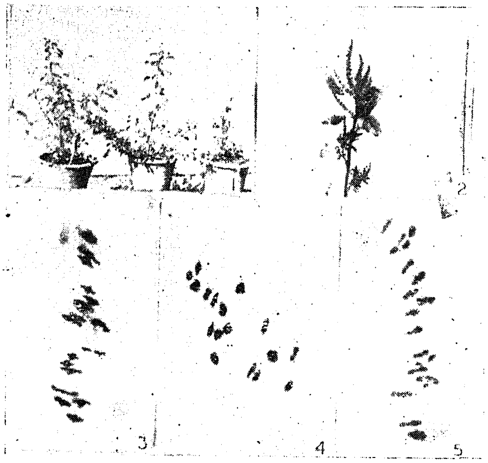
BOTH the parental species are wild and belong to the section Blitopsis of the genus *Amaranthus*. While *A. viridis* L. is a worldwide weed, *A. graecizans* L. occurs in cultivated fields and appears to have been naturalised from tropical America[2]. Their tender tips are often used as pot herb, although these are not valued as much as those from *A. tricolor* L. and *A. lividus* L. A number of naturally occurring interspecific hybrids have been reported from this section[1], but only some have been cytologically investigated[3-4]. However, the present hybrid has been investigated for the first time and some interesting morphological and cytological facts are being reported here.

Seeds of *A. graecizans* were collected from Coimbatore and those of *A. viridis* from Lucknow. The particular form of the latter taxon used in this study conformed to the description of the typical *A. viridis* in the taxonomically important features of perianth and utricle, but differed slightly in colour and leaf shape. These differences fall within the overall range of the species and its taxonomic identity was confirmed by the authorities at Kew.

The two species do not seem to be very compatible. Hybrids were raised after removing young unopened male flowers from each glomerule of *A. graecizans*, followed by heavy dusting with *A. viridis* pollen. Even so, more than half of the progeny comprised of the maternal parent resulting from the self-pollination due to subsequent emergence of male flowers in the glomerule, which could not be removed without damaging the whole glomerule. Cytological studies were carried out from the pollen mother cells for which the young inflorescences were fixed in Carnoy's fluid to which a few drops of 45% acetic acid saturated with iron acetate had been added. After 48 hours the buds were squashed in 1% acetocarmine.

Morphologically, the hybrid was intermediate in branching pattern, leaf shape and colour with an overall dominance of the female parent (*A. graecizans*) except for the small 2–4 cm long terminal inflorescence (Fig. 2), which is absent in *A. graecizans* but present in *A. viridis*. The flowers were normal but their number was considerably reduced to 3 to 7 in each glomerule in comparison to 23 to 38 and 7 to 10 in female and male parents respectively. Very few male flowers were observed in the hybrid. Some glomerules were even completely devoid of them.

In both the species 17 bivalents were observed at diakinesis and metaphase I (Figs. 3–4). In *A. graecizans* one of the bivalents was seen to disjoin early in most of the cells, a feature common to many species of the genus[5]. The bivalents had one or two chiasmata. Anaphase I was regular in both the taxa. Subsequent stages of meiosis were also regular resulting in normal pollen and seed setting.



FIGS. 1–5. Fig. 1. (from l to r). *Amaranthus graecizans*, F_1 hybrid and *A. viridis*. Fig. 2. Terminal portion of a branch of the hybrid. Figs. 3–5. Metaphase I. 17 II. *A. graecizans*. *A. viridis* and F_1 *A. graecizans* × *A. viridis* respectively. (All, × 800).

Like the parents, the hybrid also showed 17 bivalents at diakinesis and metaphase I (Fig. 5). Anaphase I, however, was characterised by late disjunction of 1 to 5 bivalents. Ultimately the components reach their poles and further course is apparently normal. However, there is complete pollen and seed sterility.

Khoshoo and Pal[3] and Pal and Khoshoo[4-5] observed translocations involving 4 to 14 chromosomes in the four different interspecific hybrids, studied by them from the section Blitopsis to which the parents of the present hybrid belong. On the basis of their observations, they concluded that chromosomal translocations appeared to be an important factor underlying species differentiation in this section of the genus. However, in the interspecific hybrid reported here only bivalents were observed and no evidence of chromosomal association or translocation was found. The situation therefore appears to be akin to what has been reported by Pal and Khoshoo[4] in section *Amaranthus* of the genus where there is complete bivalent pairing in the hybrids and chromosomal differentiation is limited only to small segments (cryptic structural hybridity *sensu* Stebbins[6]) which do not in any way impair bivalent pairing but result in complete pollen and seed sterility.

The author is grateful to Dr. T. N. Khoshoo, Deputy Director-in-charge, National Botanic Gardens, for his guidance and facilities and sincere thanks are due to Dr. M. Pal for the material and to Mr. T. K. Sharma, for the illustrations. The present study was carried out under the PL-480 grant FG-In-511.

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1. Aellen, P., *Die Amaranthaceen Mittleuropas*, Carl Hanser, Verlag, Munchen, 1961.
2. Jepson, W. L., *A Manual of Flowering Plants of California*, University of California Press, Berkeley and Los Angeles, 1951.
3. Khoshoo, T. N. and Pal, M., *Chr. Today*, 1972, 3, 259.
4. Pal, M. and Khoshoo, T. N., *Theoret. Appl. Genet.*, 1973 *a*, 43, 242.
5. —, *Ibid.*, 1973 *b*, 43, 343.
6. Stebbins, G. L., *Variation and Evolution in Plants*, New York, Columbia Univ. Press, 1950.

A TWIN GRAIN MUTANT IN BARLEY

A TWIN grain mutant was observed in the segregating population of a cross between a 6-rowed barley variety K 24 and a 2 rowed exotic barley culture EB 772. In this mutant most of the central spikelets were doubled and the extra spikelet was covering the central one and produced grains. (Figs. 1–2, 3).

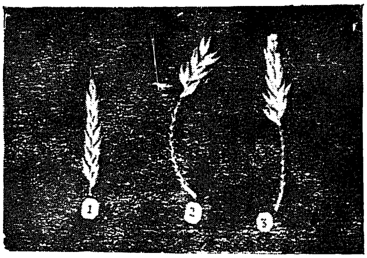

FIG. 1. Samples of normal and mutant spike where lateral grains are removed. 1, normal; 2 and 3, Twin grains.

Critical observation revealed that the rachilla producing the central spikelet was branched and produced the extra spikelet. Pollen and ovule fertility were normal, however, some of the extra spikelets produced no grains.

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SHORT SCIENTIFIC NOTES

Extractive Spectrophotometric Determination of Ce(IV) with *p*-Anisidine

A new and simple extractive spectrophotometric method for the determination of Ce(IV) with *p*-anisidine is worked out. The radish violet complex has an absorbance maximum at 510 nm. at pH 2·0. The colour of the complex is not stable in aqueous medium for a long time, however, if extracted with organic solvent like isoamyl alcohol, isobutyl alcohol, chloroform, etc. the colour is stable for several days. Beer's law is valid up to 20 ppm. As low as 0·5 ppm of Ce(IV) can be estimated. Molar absorptivity is 1366. Sandell's sensitivity calculated by using formula $ss = 10^3$ AC min. is 0·1 μg/cm^2. Composition of the complex according to Job's method of continuous variation is 1:2. The presence of Cu^{++}, Ni^{++}, Co^{++}, Mg^{++}, Mn^{++}, other lanthanides, Cl$^-$, Br$^-$, NO_3^- in tenfold excess can be tolerated in the estimation of Ce(IV).

Recommended Procedure

An aliquot of the solution containing up to 20 μg of cerium is mixed with 5 ml of the reagent (1%) and the pH of the mixture is adjusted to 2 using KCl–HCl buffer. The solution is diluted to 25 ml and shaken with 10 ml of isoamyl alcohol in a separating funnel. After separation of the two layers, the absorbance of the organic layer is measured at 510 nm after appropriate dilution.

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Chemical Studies on *Bauhinia racemosa*

The stem bark of *Bauhinia racemosa* (Fam.: Leguminosae) has been used in the indigenous system of medicine against the infection of malaria, diarrhoea and dysentery[1]. Since no chemical work on this plant species is on record, a preliminary chemical study on its stem bark has been presented.

Dried and pulverised stem bark was thoroughly extracted with petroleum ether (60°–80°) in a soxhlet. The extract was concentrated and subjected to chromatographic resolution over Brockmann alumina. Elution of the column with petroleum ether (60°–80°) furnished a white waxy mass crystallising from acetone as white shinning flakes, m.p. 61·0°; found C, 85·08; H, 14·24; calculated for $C_{28}H_{58}$, C, 85·56; H, 14·77; NMR ($CDCl_3$) characteristic of long chain alkanes, (δ 0·86, *t*, for two terminal methyl groups and δ 1·30, envelope, for polymethylenes). The compound is probably octacosane (reported m.p. 61·5°)[2].

Benzene eluates upon concentration and cooling gave a white residue which recrystallised from absolute ethanol as needles, m.p. 195°, gave Liebermann Burchard test positive for triterpenes; I.R. ν_{max} (cm^{-1}) 3300–3400 (– OH). With acetic anhydride and triethyl amine at room temperature it furnished an acetate, m.p. 236–237° giving M^+ at m/e 468 and other prominent fragments at m/e 453, 408, 393, 218 (100%) 203 and 189. The mass fragmentation pattern is in agreement with that of β-amyrin acetate. Thus the parent compound was characterised as β-amyrin and itsi dentity confirmed by its m.m.p, co-TLC studies, I.R. and the m.m.p. of the derivatives with those of the corresponding authentic samples. Further elution of the column with benzene, chloroform (3 : 1) yielded β-sitosterol, m.p. 135–136° which gave an acetate, m.p. 126–127° and a benzoate, m.p. 144–145° and its identity wae confirmed by m.m.p., co-TLC and superimposable I.R. with that of its authentic samples.

Thanks are due to Dr. A. B. Ray of the Department of Medicinal Chemistry, Banaras Hindu University, for supplying an authentic sample of β-amyrin.

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RATTAN LAL KHOSA.

1. Chopra, R. N., Nayar, S. L. and Chopra, I. C., *Glossary of Indian Medicinal Plants*, C.S.I.R., New Delhi, 1956, p. 34.
2. Weast, Robert, C. and Selby Samuel, M., *Handbook of Chemisry and Physics*, The Chemical Rubber Co., Cleveland, Ohio, 48th Ed., 1967–68, C–435.

The Possibility of Cultivation of *Solanum khasianum* by Stem Cuttings

In comparison to the propagation by seeds[1-2], the writers found that *S. khasianum* plants can be successfully propagated by stem cuttings with apical bud and leaves remaining intact. If the cuttings are made in the months of July–September, 80-90% success is achieved. The root initiation is found to occur after 10–12 days from the date of cutting. Leaves, branches, vegetative buds, floral buds, fruits and alkaloid contents are found to be greater in plants grown from August-cuttings in comparison to the plants grown from seeds. The greater number of fruits and correspondingly higher solasodine content occurs possibly due to the repeated early branching in case of plants grown from cuttings.

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(West Bengal), *June* 21, 1976.

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1. Maity, P. C., Mukherjee, S., Mathew, R. and Henry, A. N., *Curr. Sci.*, 1964, **33**, 730.
2. Sainy, A. D. and Biswas, R. C., *Ind. J. Plant Physiol.*, 1967, **10** (1) 36.

Two New Leaf Spot Diseases of *Crotalaria juncea* L Caused by *Pleospora infectoria* Fuckel and *Phoma glomerata* Corda.

During the survey (September and October 1973) of *Crotalaria juncea* in cultivated field, a number of leaves were found infected with typical spots. Two fungi were isolated from infected leaves using potato dextrose agar (P.D.A.) medium at 30° C. In the case of one, the spots were pale yellow turning black and 7–15 mm in dia. The fungus was identified as *Pleospora infectoria* Fuckel by standard methods. In the case of other, the spots appeared as yellow turning to brown in colour and 3–5 mm in dia. The culture was identified as *Phoma glomerata* Corda.

The spore suspensions of the two fungi in sterile water were sprayed on healthy plants which showed typical symptoms over the leaves after 7 and 10 days, respectively. Reisolations from the leaves of artificially inoculated plants yielded the same pathogens.

The fungus *Pleospora infectoria* and *Phoma glomerata* causing leaf spot diseases of *Crotalaria juncea* are the first records. The cultures were identified from and deposited in CMI, England (IMI 197814 and IMI 197817).

The authors express their grateful thanks to Dr. M. N. Gupta, Dr. K. D. Sharma and Dr. C. P. Agarwal for facilities and suggestions and the Director, CMI, England, for confirming the identity of fungi.

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School of Studies in Botany, R. K. S. CHAUHAN.
Jiwaji University, Gwalior,
August 7, 1976.

Allamanda cathartica, A New Host of *Colletotrichum gloeosporioides*

A leaf blight disease was observed on *Allamanda cathartica* Linn., a common ornamental plant, in the Allahabad University Campus and in adjacent regions during January 1976. Isolations were made from the surface sterilized diseased spots and the fungus was identified as *Colletotrichum gloeosporioides* Penz., the conidial stage of *Glomerella singulata* (Ston.) Spauld. and Schrenk.

Pathogenicity trials were carried out by spraying the conidial suspension on healthy leaves which developed typical disease symptoms.

The authors are grateful to Dr. S. N. Bhargava for his valuable suggestions and to Prof. D. D. Pant, Head of the Department of Botany, for providing necessary laboratory facilities.

Botany Department, D. N. SHUKLA.
University of Allahabad, A. P. SINGH.
Allahabad, *July* 1, 1976.

'CH-I' A New 'Plant Type' in Castor (*Ricinus communis* L.)

During *Kharif*, 1969, a very dwarf plant was located in Gujarat Castor Hybrid–3 in the crop grown in Haryana Agricultural University, Hissar. This extremely dwarf plant was very impressive by its bushy appearance. This plant was, therefore, selfed. At flowering, it was observed that its spikes had only 10–15% male flowers. The seed setting on selfing was good. The progeny of this selfed plant was grown during *Kharif*, 1970. Some plants were again selected and selfed. In a few seasons, a pure genotype was obtained and tested in yield evaluation trials.

Characteristics

CH–I, the new variety, is extremely dwarf (90–110 cm depending upon the soil and environment), with short internodes, leaves closely placed giving 'rosette' appearance in vegetative stage, bushy appearance becoming apparent in 35 days, foliage dark green and crowdy and profuse branches, the branches bearing 5–8 spikes. The spikes on branches are quite comparable with the spikes on the main shoot. These spikes mature synchronously in 105–115 days. There are 75–100 capsules per spike with about three small seeds per capsule, seeds contain 48% oil.

Results of field trials indicated that CH–I, the new strain, matures earlier as compared to 140–150 days in Aruna (NPH–I) and 270–285 days in PC No. I. It is very dwarf when compared to 340–350 cm height in Aruna and 400–425 cm in PC No. 1. The yield (16 Q/ha) is at par with Aruna (17 Q/ha) and lower than PC No. 1 (20 Q/ha) under irrigated conditions. The oil content is more or less the same in all the varieties.

Due to the early maturity dwarfishness and desirable plant type, CH–I, would fit well in double cropping which was not possible with other varieties in castor. With a maturity of 110–120 days, CH–I can be grown as a pure *Kharif* crop to be followed by any *Rabi* crop.

Department of Plant Breeding, T. P. YADAVA.
Haryana Agricultural University, HARI SINGH.
Hissar 125 004, India, C. K. YADAV.
January 27, 1976.

Purification of Petunia Mottle Virus

The petunia mottle virus (PMV) is relatively unstable in most of routine purification procedures. The following two methods were found to be satisfactory.

Leaves of young *Petunia hybrida* Vilm. plants, inoculated 10 days earlier with PMV, were freezed and 100 g material was homogenised in 50 ml distilled water containing 0·75 g sodium ascorbate (Gooding[2]). The slurry was squeezed through two layers of cheese cloth, and 6–8% v/v *n*-butanol was added and a low speed of centrifugation was given. The pellet was discarded and supernatant was concentrated by 3 alternate cycles of low and high speed centrifugation by resuspending the pellets obtained at high speed in 0·1 M borate buffer pH, 8·0. The final pellet was dissolved in 2 ml of 0·1 M borate buffer, pH 8·0 and was subjected to sucrose density gradient (Brakke[1]) and an absorbance band was conserved after 4½ hours run at 24,000 rpm.

In the second procedure 100 g of infected petunia leaves were macerated with 150 ml of 0·1 M borate buffer, pH 8·0 (Francki[3]). The sap was squeezed through two layers of cheese cloth and was freezed overnight. After freezing, the sap was thawed and centrifuged using low speed of 10,000 rpm for 10 minutes. Granular ammonium sulphate was added in proportion of 15 g/100 ml extract and was stirred for 6–8 hours in magnetic stirrer and then low speed centrifugation was given. The supernatant fluid was concentrated by two cycles of high and low speed centrifugation. The final pellets obtained were resuspended in 0·1 M buffer, pH 8·0 and the virus solution was subjected to sucrose density gradient centrifugation (Brakke[1]) and an absorbance band was observed after 4½ hours run at 24,000 rpm.

The infectivity of the virus after each step in both the purification procedures was asseyed on *Chenopodium amaranticolor* Coste and Reyn., a local lesion host for PMV. The results are given in Table I,

TABLE I

Infectivity of petunia mottle virus when purified with two different methods (a—sodium ascorbate and butanol method and b—ammonium sulphate method)

Stages of purification	Number of local lesions showing infectivity of different dilutions of PMV				
	Undiluted	10^{-1}	10^{-2}	10^{-3}	10^{-4}
(*a*) *Sodium ascorbate and butanol method*:					
Clarified extract after treatment of butanol	78	54	39	20	0
After I cycle	64	42	24	14	0
After II cycle	66	40	26	12	0
After III cycle	62	34	20	8	0
After density gradient centrifugation	48	22	8	4	0
(*b*) *Ammonium sulphate method*:					
Clarified extract after treatment of ammonium sulphate	82	60	42	18	0
After I cycle	60	48	34	14	0
After II cycle	54	42	28	10	0
After density gradient centrifugation	46	32	20	6	0

Values are average of numbers of lesions per leaf. Three plants having 6 leaves each were used.

Department of Botany,
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Aligarh 202 001, India, *June* 16, 1976.

QAMAR A. NAQVI
K. MAHMOOD.

1. Brakke, M. K., *Adv. Virus Res.*, 1960, **7**, 193.
2. Gooding, G. V., *Phytopathology*, 1963, **53**, 475.
3. Francki, R. I. B., In: *Principles and Techniques in Plant Virology* (Eds. C. I. Kado and H. O. Agrawal), van Nostrand Reinhold Co., 1972, p. 295.

New Record of *Euproctis semisignata* Walker (Lymantriidae: Lepidoptera) as a Serious Pest of Coconut Inflorescence

During the course of a breeding programme for the production of Tall × Dwarf hybrid nuts in the Trichur District, *Euproctis semisignata* Walker was observed for the first time as a serious pest of female flowers and tender buttons aged upto three months. The pest infestation was observed during March–May,

TABLE I

Developmental period (days) of various stages of E. semisignata

Incubation period of eggs*		Larval period**		Pupal period**		Meteorological conditions			
						Temperature °C		RH%	
range	mean	range	mean	range	mean	Min.	Max.	07·25 hrs	14·25 hrs
5–9	6·50	19–35	24·86	7–10	8·10	23·70	35·00	88·23	55·72

* Based on 10 egg masses. ** Based on 15 individuals.

in the coastal regions of Cranganore and in isolated patches around Irinjalakuda. *E. semisignata* was reported to be of common occurrence in India by Lefroy (1909).

The larvae bore into the female flowers and the developing buttons through the stigmatic ends and fed on the nucellar tissue. In open inflorescence the extent of damage was 5–10%. Under bagged conditions the infestation was relatively higher, the range being from 25 to 50%.

The adult moth is dusky yellowish with a median transverse comma-like broad black band across the forewings. Eggs are laid in masses of 15–30 (mean = 22: $n = 10$) on main and subsidiary rachis of the spadix towards the basal regions and covered over with anal hairs. The duration of development of various stages are indicated in Table I.

The total larval period lasts for about 25 days. Pupation takes place on the rachis. The pupa is obtect, characterised by the presence of larval verrucae on the abdominal segments and are enclosed in silken cocoons containing urticating hairs. The pupal period lasts for 7–10 days.

The authors are thankful to Dr. G. S. Arora, Zoologist, Zoological Survey of India, Calcutta-12, for establishing the identity of the pest and to Sri. K. M. Thomas, Assistant Director, Department of Agriculture, Kerala, for the help rendered in making field collections.

College of Horticulture,
Mannuthy 680 651, Trichur,
Kerala, *June* 4, 1976.

C. C. ABRAHAM.
K. S. REMAMONY.

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1. Lefroy, H. M., *Indian Insect Life*, 1909, p. 461.

A New Record of *Pythium* from India

During a survey of Saprolegniaceous fungi occurring in certain soils of Gorakhpur, a few members of Pythiaceae (Order: Peronosporales) were also found to be distributed in sandy soils of St. Andrew's College campus having organic content 4·0% and moisture content 7·5%. One species of *Pythium* with conspicuous ornamented oogonial walls was isolated from this locality. Soil sampling was done on the lines suggested by Dick and Newby[1] and unifungal, bacteria-free cultures were raised on sterilized hemp-seed halves at 25–28° C.

The isolate was identified with the help of the monograph by Middleton[2] as *P. echinulatum* Mathews.

The work was supported by a grant from the C.S.I.R., New Delhi. The authors are thankful to Dr. Y. B. Singh, Principal, for providing laboratory facilities and encouragement.

Department of Botany,
St. Andrew's College,
Gorakhpur 273 001,
May 28, 1976.

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1. Dick, M. W. and Newby, H. V., *J. Ecol.*, 1961, **49**, 403.
2. Middleton, J. T., "The taxonomy, host range and geographic distribution of the genus *Pythium*," *Mem. Torrey Bot. Club*, 1943.

REVIEWS AND NOTICES OF BOOKS

Standards, Specifications and Quality Requirements for Processed Casaava Products. By Jean S. Ingram. (Tropical Products, Inst., 56/62, Gray's Inn Road, London, WC IX BLU), 1975. Pp. v + 26. Price £ 0.70.

This is an authoritative monograph published by a British Governmental organisation particularly concerned with quality control and prescription of specification for commodities imported from tropical countries. The monograph details the standards and specifications prescribed by governuental authorities or requirements laid down by unofficial or trade organizations of several importing and exporting countries (India, Thailand, Malaysia, Sri Lanka, Singapore, Malagasy, Tanzania, USA and UK), for various tapioca products, such as chips, pellets, 'sago' (pearls), whole flour, starch and fermented preparations (like 'Gari' of West Africa), meant for either human foods or livestock feeds or industrial purposes. The background information provided for these specifications describes the various products and critically discusses the related quality parameters along with world production and import-export data. This brief review is well documented by references to scientific and official journals or trade publications. For years to come, the monograph is bound to prove a valuable reference source on the quality aspects of processed tapioca or cassava products.

M. V. L. Rao.

Simulation of the Fluctuations of the Grey Larch Bud Moth. By J. van den Bos and R. Rabbinge. (Centre for Agricultural Publishing and Documentation. P.O. Box 4. Wageningen, The Netherlands), 1976. Pp. 83. Price F 16·80.

The study of the population dynamics of insects is largely of interest to entomologists and ecologists but has practical value for all those connected with pest control in agriculture and public health. If the various biotic and abiotic factors that govern population fluctuations of an insect pest can be quantified, correlated and interpreted with the help of a simulation model, this may prove useful in forecasting pest outbreaks and planning and organising control measures. In some technologically advanced countries computers are being used in the study of insect populations.

The book under review deals with the development of a simulation model for the population dynamics of the grey larch bud moth, *Zeiraphera diniana*, in the Upper Engadin Valley of South-East Switzerland. After briefly describing the biolopy and ecology of the pest the authors analyse the influence and interaction of different factors, such as fecundity, egg viability, larval and pupal mortality, longevity, parasitism, diseases, damage to the tree and the loss of food quality and temperature that affect the larch bud moth populations. Using this analysis a simulation model has been designed and presented, with several diagrams and a computer programme written in CSMP. The results are discussed and a sensitivity analysis is also provided for evaluating the relative significance of the several inputs and parameters that have gone into the design of the model.

The authors have admittedly ignored, over-simplified or arbitrarily considered some important factors and are therefore aware of the limitations of their model. They stress the need for further experiments and field studies to verify their conclusions.

The monograph is handy and well produced. It is one of a series on computer simulation in agriculture and its supporting sciences.

T. Sankaran.

Space Structures—Their Harmony and Counterpoint. By Arthur L. Loeb. (Addison Wesley Publishing Co., Reading, Mass.), 1976. Pp: xviii + 169. Price : Cloth binding $ 19.50 ; paper binding $ 9.50.

The book under review deals both with the topological and with symmetry aspects of space structures, this being the author's way of describing a system of points connected by lines along with the faces and cells which they generate. The treatment will appeal to crystallographers, specially those interested in structural and decorative design and, in general, to the geometrically inclined. The author starts with the basic Euler-Schläfli relation connecting the number of points, lines, planes and cells. The full power of this seemingly simple relation is revealed in the systematic and exhaustive enumeration of regular and semiregular figures in two dimensions. There is a strong emphasis on the mutual relationships of the various patterns, the basic tools being, truncation and duality (replacement of faces by vertices). These in turn are unified by introducing the concepts of edge truncation and edge stellation.

The later part of the book deals with space filling figures and Dirichlet domains (familiarly known to solid state physicists as Wigner-Seitz cells). Here the emphasis is on specific cases with cubic, tetragonal, or orthorhombic symmetry rather than the general theory, and again the strong point of the book is in bringing relationships between different figures to light. No wonder the author describes his aim as one of creating a 'structure of structures'. This section and the accompanying photographs provide a stimulus to thinking in three dimensions, an art practised by crystallographers, although it is now under siege by the one dimensional science of computer programming. Books like this one may yet keep the former alive.

Rajaram Nityananda.

ANNOUNCEMENTS

Second 'Oriental Entomology' Symposium

The Second 'Oriental Entomology' Symposium is planned to be held at Madras in March, 1977. Further information may be obtained from the Convener, Dr. T. N. Ananthakrishnan, Entomology Research Unit, Loyola College, Madras 600 034.

Genetic Studies on Mosquitoes

A Symposium on "Genetic Studies on Mosquitoes" will be organized during the Third Week of November. 1976 under the auspices of the Bangalore University with the support of the University Grants Commission.

Papers on the following topics will be presented at the Symposium: 1. Cytogenetics, 2. Formal Genetics, 3. Genetics of Insecticide Resistance, 4. Bio-Chemical Genetics, 5. Physiological Genetics, 6. Genetic Effects of Radiation and Chemosterilants, 7. Mosquitoes and Evolution, 8, Ecological Studies and Their Bearing on Vector Control, 9. Vector Control Mechanisms (*a*) Chemical, (*b*) Biological, (*c*) Genetical, 10. Mosquito Borne Diseases.

The participants are requested to write to the Convener. Dr. B. N. Chowdaiah, Department of Zoology, Bangalore University, Bangalore 560 001.

Indian Science Congress Association

The 64th Session of the Science Congress will be held at Bhubaneswar from January 3 to 7, 1977.

The Focal Theme of 'Survey, Conservation and Utilisation of Resources' will be discussed in this session under the joint auspices of the Utkal University and the Orissa University of Agriculture and Technology.

This Focal Theme would be introduced by the President, Dr. H. N. Sethna, through his Presidential Address to be delivered on January 3, and specific aspects of the theme would be discussed by the thirteen Sections of the Science Congress covering all the major disciplines as also its two Statutory Committees, namely, the Committee on Science and Its Social Relations and the Committee on Science and Economic Development, and the Forum on Home Science and Nutrition.

Details regarding the subjects discussed at various sections of the Science Congress can be had from the General Secretaries, Indian Science Congress Association, 14 Dr. Biresh Guha Street, Calcutta-17.

6120/76. Published by S. R. S. Sastry, Manager, Current Science Association, Bangalore-560006
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2-PICOLINE-N-OXIDE COMPLEXES OF RARE-EARTH CHLORIDES

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ABSTRACT

2-Picoline-N-Oxide (2-PicNO) complexes of seven rare-earth chlorides of the general formula M (2-PicNO)$_n$ Cl$_3$ where $n = 4$ for M = La or Pr; and $n = 5$ for M = Nd, Sm, Tb, Er or Y have been prepared and characterized. Ligand is coordinated through oxygen of the N–O group and the chloride ions are non-ionic.

INTRODUCTION

IN continuation of our recent studies on 2-PicNO complexes of rare-earth bromides[1], iodides[2], nitrates[3] and perchlorates[5], the preparation and characterization of its complexes with rare-earth chlorides is undertaken.

EXPERIMENTAL

Preparation of the complexes:—One g of the ligand was added to about 0·5 g of the hydrated rare-earth chloride. The mixture was triturated and warmed on a water bath for about 15 minutes. The fine crystalline product obtained after cooling was suspended in acetone in which only the free ligand is quite soluble. The product was filtered through a sintered crucible, washed well with acetone and dried in a desiccator over calcium chloride under reduced pressure.

Analyses: The metal content of the complexes was estimated by EDTA titrations using xylenol orange as indicator; chloride was estimated by Volhard's method and 2-PicNO spectrophotometrically in aqueous solutions at 254 nm using the calibration curve method. The infrared, and conductivity measurements were carried out as described earlier[5].

RESULTS AND DISCUSSION

Results of analyses (Table I) conform to the general formula M (2-PicNO)$_n$ Cl$_3$ where $n = 4$ for M = La or Pr; and $n = 5$ for M = Nd, Sm, Tb, Er or Y. Unlike in the corresponding bromide, iodide, nitrate and perchlorate complexes, the metal ligand ratio changes as one goes from lighter to heavier lanthanides. The stoichiometry of the complexes is different from that of the corresponding pyridine-N-oxide complexes[6]. While eight molecules of PyO are coordinated to all the rare-earth metal ions only four or five ligand molecules are coordinated in the present case. This is due to the steric effect of the methyl group at 2-position in the aromatic ring of PyO. It is interesting to observe that the reduction in the number of ligand molecules coordinated to rare-earth ions, as one goes from PyO to 2-PicNO complexes, is more in rare-earth halide complexes (3 or 4 molecules) than in the perchlorate complexes (1 molecule).

The complexes are hygroscopic, insoluble in non-polar solvents and acetone but soluble in alcohols, DMF and DMSO. While the complexes of La, Pr, Nd and Sm are insoluble in acetonitrile, those of Tb, Er and Y are slightly soluble. Though all the complexes

TABLE I

Analytical and conductance data for the complexes

Compound	Metal (%)		Chloride (%)		2-PicNO (%)		Molar conductance (ohm^{-1} cm^2 mole^{-1}) in	
	Calc.	Found	Calc.	Found	Calc.	Found	Methanol	Acetonitrile
La (2-PicNO)$_4$ Cl$_3$	20·39	20·52	15·62	15·46	64·03	64·20	126·7	..
Pr (2-PicNO)$_4$ Cl$_3$	20·61	20·67	15·57	15·61	63·84	63·79	126·0	..
Nd (2-PicNO)$_5$ Cl$_3$	18·12	18·04	13·37	13·52	68·52	68·43	127·6	..
Sm (2-PicNO)$_5$ Cl$_3$	18·74	18·59	13·26	13·40	67·98	68·02	129·7	..
Tb (2-PicNO)$_5$ Cl$_3$	19·59	19·45	13·12	13·02	67·28	67·50	130·8	34·64
Er (2-PicNO)$_5$ Cl$_3$	20·42	20·40	12·99	12·82	66·59	66·37	124·1	26·48
Y (2-PicNO)$_5$ Cl$_3$	12·00	11·96	14·36	14·15	73·64	73·81	124·0	33·56

are soluble in chloroform, they separate out subsequently on standing except for the Er complex which does not separate from the solution. The colours of the complexes are similar to those of the corresponding rare-earth salts.

The Tb, Er and Y complexes behave as non-electrolytes in acetonitrile suggesting that all the chloride ions are coordinated to the metal. Hence a coordination number of seven for lighter lanthanides (La and Pr), and eight for heavier lanthanides and yttrium can be postulated. An increase in the coordination number on going from lighter to heavier lanthanides can be explained as follows. On going from La to Lu, the size of the tirvalent lanthanide ion decreases and hence steric effect will influence the coordination number of the lanthanides leading to a lower coordination number for the heavier lanthanide. At the same time the ionic potential of the trivalent lanthanides increases on going from La to Lu. The heavier lanthanides attracting more number of ligands show that the influence of the second factor is dominant in the present case.

The infrared spectra of the complexes are similar showing no significant dependence on the central meta ion. The N–O stretching vibration of the ligand appearing at about 1255 cm^{-1} is shifted to about 1234 cm^{-1} in the complexes indicating bonding of the ligand through oxygen of the N–O group. The N–O bending vibration occurring at 855 cm^{-1} in the free ligand remains unshifted. The C–H out-of-plane vibration appearing at 775 cm^{-1} is shifted to slightly higher frequencies due to decrease in the electron density in the aromatic ring consequent upon the formation of the metal to ligand bond.

The extent of ν_{N-O} shift in the complexes which is a measure of the metal-oxygen bond strength is almost the same in rare-earth chloride, bromide and iodide complexes of 2-PicNO.

ACKNOWLEDGEMENT

One of the authors (P. V. S.) thanks C.S.I.R., India, and the authorities of the Indian Institute of Science for the financial assistance.

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1. Sivapullaiah, P. V. and Soundararajan, S., *J. Inorg. Nucl. Chem.*, 1976, **38**, 1402.
2. Ramakrishnan, L. and Soundararajan, S., Unpublished results.
3. — and —, *Monatsh Chem.* (in press).
4. Koppikar, D. K. and Soundararajan, S., *Inorg. Nucl. Chem. Lett.*, 1976, **12**, 459.
5. Sivapullaiah, P. V. and Soundararajan, S., *Curr. Sci.*, 1975, **44**, 374.
6. — and —, Unpublished results.

PHYSICOCHEMICAL INVESTIGATION OF SOME TRIVALENT METAL-CHELATES OF *o*-(α-2-OXOPROPYLBENZYLIDENEIMINO) BENZENE SULPHONIC ACID

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A SURVEY of the literature[1] has revealed tha no work has been done on the rare-earth chelates of *o*-(α-2-oxopropylbenzylideneimino) benzene sulphonic acid (H_2OB). Hence the physicochemical studies of the metal-chelates of La(III), Ce(III), Pr(III), Nd(III), Sm(III) and Gd(III) with H_2OB have been carried out using potentiometric, magnetic and spectral techniques and their stoichiometry and structures have been established.

Potentiometric titrations of the mixtures containing (A) perchloric acid alone, (B) perchloric acid and the ligand (H_2OB) and (C) perchloric acid, ligand and metal-ion solution have been carried out by the method of Irving and Rossotti[2].

The mean values of dissociation constants (log K_1^H and log K_2^H) as obtained by interpolation at half $\bar{n}$ values method[3] and interpolation of various $\bar{n}$ values method[4] have been found to be 9·755 and 3·030 at 25° and 9·355 and 2·880 at 35°, respectively. The formation curves for the metal-ligand system attain maxima at $\bar{n} > 1 \cdot 5$ which indicates that 1 : 1 and 1 : 2 chelates are formed. Since in all the cases $K_1/K_2 > 10^{2.5}$, the method of interpolation at half $\bar{n}$ values is applicable besides the methods of successive approximation and correction term[4]. The values of the stability constants as obtained from these methods are in good agreement and the average values thus obtained are shown in Table I. Thus the stability of the chelates increases, with decreasing ionic size of the metal-ions, *i.e.*, La (III) < Ce (III) < Pr (III) < Nd (III) < Sm (III) < Gd (III). The values of their thermodynamic parameters such as free energy change ($\triangle G^\circ$), enthalpy change ($\triangle H^\circ$), entropy change ($\triangle S^\circ$) and the energy of activation (E) have also been included in Table I. The more negative values of $\triangle G^\circ$ at 35° C than at 25° C and the positive values of $\triangle H^\circ$ in all the cases suggest the endothermic nature of these reactions. The positive values of $\triangle S^\circ$ indicate that the entropy term is favourable for their formation. The positive values of energy of activation further confirm the endothermic nature of the reactions.

TABLE I

Stability constants at 25° and overall thermodynamic parameters at 30°

Metal-chelates	Stability constants	$-\Delta G°$ K.Cals/mole	$\Delta H°$ K.Cals/mole	$\Delta S°$ Cals/deg./mole	E K.Cals/mole
La (III)					
log K_1	6·40 (6·79)	14·35 (15·47)	47·04	204·5	47·04
log K_2	4·13 (4·19)				
Ce (III)					
log K_1	7·55 (7·60)	16·95 (18·03)	45·36	207·4	45·36
log K_2	4·88 (5·19)				
Pr (III)					
log K_1	8·10 (8·20)	18·18 (19·46)	53·76	239·5	53·76
log K_2	5·23 (5·61)				
Nd (III)					
log K_1	8·49 (8·62)	19·65 (20·97)	55·46	205·1	55·46
log K_2	5·92 (6·25)				
Sm (III)					
log K_1	9·00 (9·10)	21·03 (22·46)	60·06	270·1	60·06
log K_2	6·42 (6·83)				
Gd (III)					
log K_1	9·22 (9·35)	22·07 (23·34)	53·34	251·0	53·34
log K_2	6·96 (7·21)				

The values in parenthesis are at 35°C.

Synthesis of rare-earth chelates.—To an ethanolic solution of H_2OB (0·022 M), a solution of the rare-earth metal-nitrate (0·02 M) in 80% ethanol was added and the mixture stirred. Dilute ammonia was gradually added to this mixture until flocculent greenish yellow mass was formed. It was filtered, washed with hot ethanol, dried in air and preserved in a desiccator. The yields were found approximately 75%. Satisfactory carbon, hydrogen, nitrogen, sulphur and metal analyses have been obtained for these chelates. These data suggest 1 : 2 metal-ligand stoichiometry in these compounds.

The rare-earth chelates undergo exchange reactions with EDTA in water and thus EDTA-chelates of the metal-ion of interest have also been prepared.

The rare-earth chelates decompose without melting on heating above 250° C to give their oxides at 410° C.

An infra-red study of H_2OB has shown three bands at 1170 cm^{-1}, 1680 cm^{-1} and 3600 cm^{-1} which can be assigned to the presence of sulphonic acid ($-SO_3H$) $>C=N$ and enolic $-OH$ respectively. In all the metal-chelates under study the bands at 1170 cm^{-1} and 3600 cm^{-1} could not be located which suggest their elimination due to complexation.

With the exception of La (III)-chelate which is found diamagnetic, the magnetic moments (in B.M. at 303° K) of the other chelates, as obtained by Gouy method, are: Ce (III)-(2·25), Pr (III)-(3·33), Nd (III)-(3·65), Sm (III)-(1·49) and Gd (III)-(7·85). These values are in agreement with those of the lanthanide sulphates[5]. These values suggest that the lanthanide ion in the chelates acts approximately as free-ion, as far as the f-electrons are concerned.

ACKNOWLEDGEMENT

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1. Holm, R. H., Averett (Jr.), G. W. and Chakravorty, A., *Prog. Inorg. Chem.*, 1966, **6**, 88.
2. Irving, H. and Rossotti, H. S., *J. Chem. Soc.*, 1945, p. 2904.
3. Bjerrum, J., *Metal Ammine Formation in Aqueous Solution*, P. Haase and Sons, Copenhagen, 1957, pp. 20, 114, (14 (*a*), 36 (*b*).
4. Irving, H. and Rossotti, H. S., *J. Chem. Soc.*, 1953, p. 3397.
5. Figgis, B. N. and Lewis, J., *Techniques of Inorganic Chemistry*, Interscience Publishers, Inc., New York, 1965, **4**, 137.

STUDIES ON ANTHER CULTURE

I. Induction of Androgenic Plants and Callus in *Petunia hybrida* Vilm.

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EVER since the demonstration in *Datura innoxia* by Guha and Maheshwari (1967), attempts are being made to induce androgenesis in a number of plants. Sunderland (1971) has discussed the problems and prospects of rearing pollen embryoids and their subsequent growth to maturity in several plants with special reference to the family Solanaceae. From 1971 onwards, we began experiments with the view to exploiting this technique for producing androgenic plants in horticultural crops. In *Petunia*, it was envisaged that haploids and homozygous lines obtained thereof would be of great value in a future breeding programme (Dore Swamy and Chacko, 1973). This paper highlights some of our results in producing androgenic diploid and triploid plants.

Immature flower buds measuring 0·5 to 0·8 cm were used. Representative anthers from buds of *Petunia hybrida* Vllm. were smeared with acetocarmine to determine the stage of pollen developments. Anthers from these buds contained mostly uni-nucleate pollen grains. The flower buds were washed with distilled water and sterilised with saturated chlorine water. The anthers were excised and sterilized with saturated chlorine water and rinsed several times with sterile distilled water. The sterilized anthers were planted in batches of 4-8 on nutrient agar contained in Corning culture tubes plugged with non-absorbant cotton wrapped in cheese cloth. Either the basal medium of Nitsch (1969) NM or Murashige and Skoog (1962). MSM was used. Sucrose of 2% concentration and Na_2 Fe EDTA (32 mg/l) were added to the basal media as energy and iron sources, respectively. The media were jelled with bacteriological grade agar (0·9%). Growth adjuvants such as coconut-milk, 2, 4-dichlorophenoxy acetic acid, indole acetic acid, naphthalene-acetic acid, indole butyric acid, adenine, N^6 benzyladenine, kinetin and zeatin were added individually or in combination and the media were sterilized at 1·06 kg/cm^2 for 10 to 15 minutes. After planting the anthers, the cultures were incubated at 26° ± 2°C under 12 hours light at 1500 lux. The root tips of androgenic plants were prefixed in paradichlorobenzene, fixed in acetic alcohol and stained with Feulgen for cytological studies.

Several hundreds of anthers were cultured over a period of one year. However, only 1–2% of the anthers planted on NM containing coconut milk (20% V/V) and adenine (2 ppm) produced embryoids in 3–4 weeks.

At the end of one month, these embryoids developed into well-organised plantlets. These plantlets were subcultured in the same medium for nearly 2 months when they developed several pairs of leaves and a ramifying root system (Fig. 1). Root tip squashes of androgenic plants revealed them to be triploid with $3n = 21$ chromosomes. Anthers cultured on NM supplemented with N^6 benzyladenine (1 ppm) also developed embryoids ranging from 4–6. These embryoids developed into plantlets 7–8 weeks after culture. Root tip squashes showed them to be diploid ($2n = 14$). Anthers inoculated on NM supplemented with zeatin alone (0·1 or 1 ppm) or in combination with naphthalene acetic acid (2 ppm) also gave rise to embryoids 8–10 weeks later. But 90% of these plants were misshappen with callus formation. The root system was poorly developed and successful squash preparation could not be made. On NM supplemented either with 2, 4-D (1 ppm) or low concentrations of zeatin (0·1 or 0·2 ppm) and naphthalene acetic acid (2 ppm), the

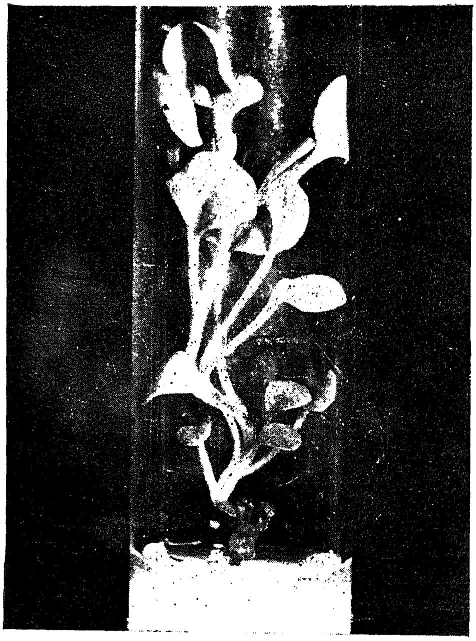

FIG. 1. Anther plant showing well-developed shoot and root system.

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anthers gave rise to actively growing callus masses. On MSM supplemented with indole butyric acid (5–10 ppm) and N^6 benzyladenine (0·5 or 1 ppm), the anthers produced callus masses which turned green. In all these instances, callus originated from the centre of the anther lobes pushing the anther walls which eventually burst open. These calli have differentiated shoot buds. Callus formation was also observed in anthers cultured on the media suggested by Engvild (1973) and Zenkteler (1973). The anther callus did not differentiate either roots or shoots even after several subcultures to media supposed to stimulate rooting.

Callus formation was noticed in anthers cultured on NM supplemented with N^6 benzyladenine (5 ppm). This callus had the appearance of a globular mass. On subculture to NM with lower concentrations of N^6 benzyladenine (2 ppm), the calli differentiated into shoots. Root differentiation was achieved (Fig. 2) in small portions of such shoots subcultured to NM supplemented with indole acetic acid (2 ppm).

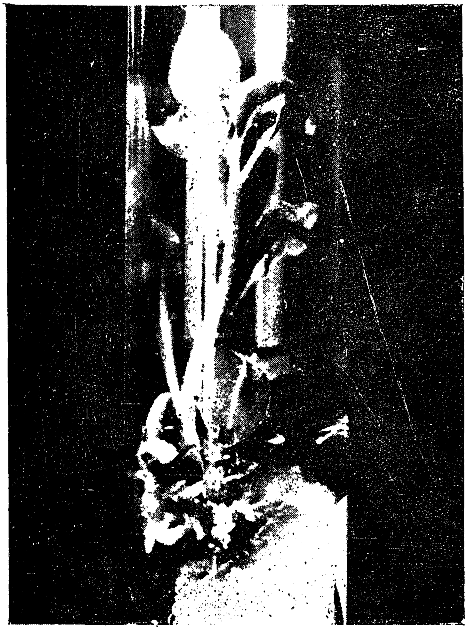

FIG. 2. Plant differentiated from anther callus.

Our results with *Petunia hybrida* suggest the necessity for a complex medium. However, even with the combination of the most favourable factors, the success is 1–2%. Raquin and Pilet (1972) have successfully obtained androgenic haploids, diploids and triploids in a hybrid of *Petunia axillaris* × *P. hybrida*. However, the plants were obtained *via* a callus. It was shown (Dore Swamy and Chacko, 1973), that plantlets and callus could be induced in *Petunia axillaris*; however, ploidy levels and origin of the plants could not be traced. Similar results were obtained by Sopory and Maheshwari (1973). Engvild (1973) obtained triploids in *Petunia axillaris* on a medium containing kinetin. We have now induced androgenic triploids in *P. hybrida*, on a medium containing coconut milk and adenine, According to Heseman (1971), in the mature trinucleate pollen grain of *Petunia*, the generative nucleus is in the double DNA (1 C) state. This prompted Raquin and Pilet (1972), to suggest that in *Petunia*, the vegetative nucleus is incapable of DNA synthesis without uniting with the generative nucleus thus giving a 3 *n* activated nucleus resulting in a triploid plant. Another possibility is that in *Petunia*, the switch to sporophytic development is incomplete and there is a slight initial tendency towards callus formation along with endomitosis and nuclear fusion. Given the proper mileu, either a plantlet or a callus may develop. However, more work needs to be done to establish the exact origin of the diploid and triploid embryoids.

Even though haploids were not produced as envisaged, the triploids and diploids produced are of value to the breeder. The fact that only reasonable success in inducing spore androgenesis in a few plants suggests that species to species differences are involved. If this is true, 'callus androgenesis', which has been more widely observed (Pandey, 1973), may be the possible tool for obtaining large number of androgenic plants in *Petunia* and others. However, successful production of haploids has been reported by Wagner and Hess (1974) and Sangwan and Norreel (1975) from pollen grains while the manuscript is being prepared.

ACKNOWLEDGEMENT

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1. Dore Swamy, R. and Chacko, E. K., *Hort. Res.*, 1973, **13**, 41.
2. Engvild, K. C., *Heriditas*, 1973, **74**, 143.
3. Guha, S. and Maheshwari, S. C., *Phytomorphology*, 1967, **17**, 454.
4. Heseman, C. U., *Theoretical Appl. Genet.*, 1971, **41**, 338.
5. Murashige, T. and Skoog, F., *Physiol. Plant.*, 1962, **15**, 473.
6. Nitsch, J. P., *Phytomorphology*, 1969. **19**, 389.
7. Pandey, K. K., *New Phytol.*, 1973, **72**, 1129.
8. Raquin, C. and Pilet, V., *C.R. Acad. Sci.*, 1972, **274**, 1019.
9. Sopory, S. K. and Maheshwari, S. C., *Curr. Sci.*, 1973, **42**, 247.
10. Sunderland, N., *Sci. Prog. Oxf.*, 1971, **59**, 527.
11. Zenktler, M., *Z. Pflanzen Physiol.*, 1973, **69**, 189.

CHANGES IN THE CHOLESTEROL CONTENT OF OVARIES AND ADRENALS DURING OVULATION INDUCED BY HOMOPLASTIC PITUITARIES IN THE GREEN FROG, *RANA HEXADACTYLA* (LESSON)

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THERE are different views with regard to the production of steroids by the amphibian ovary and the reports on the presence of steroid-dehydrogenases are contradictory[1–5]. Since the adrenocorticoids play a facilitating role during ovulation[6–7] it is believed that the maturation of oocytes and ovulation in Amphibia may be due to the steroids produced by the adrenals[1]. This investigation is an attempt to find out the relative efficiency of ovaries and adrenals in utilizing cholesterol during ovulation induced by pituitaries administered exogenously since the same may be regarded as an index of steroid production by these organs[8].

Adult frogs, *R. hexadactyla*, weighing 250 to 300 g, were collected from the vicinity of Mysore city (South India) during the breeding season and maintained separately in aerated aquaria at a room temperature of 25±1° C. They were induced to spawn with homoplastic pituitaries which were collected from gravid females of similar weight range. The pituitaries were homogenized in distilled water, in the proportion of five glands per ml and one ml of this homogenate was administered intraperitoneally into the treated ones[9]. Frogs receiving an equal volume of distilled water served as controls. The autopsy was carried out at intervals after the treatment as shown in Table I. Appropriate amounts of the ovarian and adrena tissues, which were cut and separated as far as possibl from the admixed kidney tissue, were weighed an used for the estimation of cholesterol[10]. Whil calculating the utilization of cholesterol by the ovary the amount present in the ovulated eggs was adde to that of the post-ovulatory stage and the averag was calculated. In the case of adrenals the discrepancy between the control level and that of the treate group E was taken.

It may be noted from Table I that the ovary release eggs within 1½ hours after the treatment and by abou 12 hours the ovulation is complete since no eggs ar found in the body cavity. The period from three t twelve hours constitutes the active ovulatory phase A gradual reduction in the cholesterol content of ovar occurs during the process of ovulation and it is significant by about six hours (Table II). Unlike ovarie the adrenals register a rapid decrease in their choleste content during the peak period of ovulation (group E and it is followed by a quick recovery. The per cen utilization of cholesterol is found to be 37·9 for th ovary and 53·6 for the adrenals.

Both the metabolic pathways and the enzymes involved in the synthesis of steroids by ovaries and adrenals in Amphibia are known to bear many similaritie

TABLE I

Response of ovary to the pituitary stimulation and ovulation in Rana hexadactyla

Duration of Treatment	Body weight (g) (M ± S.E.)	Weight of ovary (g) per 100 g B.W. (M±S.E.)	* Rate of release of eggs		
			Body cavity	Oviduct	Aquaria
A Control	258±34	4·2±0·9	Nil	Nil	Nil
B Treated ¾ of an hour	325±23	3·2±1·0	..	—	—
C Treated 1½ hours	305± 7	2·6±0·7	+	+	—
D Treated 3 hours	287± 7	1·7±0·2	+	++	—
E Treated 6 hours	294±20	1·4±0·1	+	++	—
F Treated 12 hours	308±12	1·2±0·1	—	++	+++

M±S.E. Mean in relation to standard error.
*+ = 1 to 10 eggs released.
++ = 10 to 50 eggs released.
+++ = 50 or more eggs.

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TABLE II

Changes in the cholesterol content of ovary, adrenals and kidney tissue during ovulation in R. hexadactyla

	Time of Autopsy			Cholesterol mg/100 g weight of tissue (M±S.E.)		
				Ovary	Adrenal	Kidney
A	Control		(8)	0·66±0·03	0·84±0·10	0·39±0·01
B	Treated	¾ of an hour	(6)	0·63±0·02	0·71±0·07	0·37±0·02
C	Treated	1½ hours	(8)	0·72±0·05	0·85±0·05	0·25±0·01
D	Treated	3 hours	(8)	0·61±0·06	0·75±0·10	0·28±0·01
E	Treated	6 hours	(10)	0·44±0·04	0·39±0·04	0·30±0·03
F	Treated	12 hours	(10)	0·43±0·04	0·81±0·11	0·36±0·04
	Ovulated eggs		(5)	0·40±0·09	..	..

Number in parenthesis indicates the number of frogs used.

	Ovary		Adrenal		Kidney	
A *vs* C	$t = 1{\cdot}016$	$P < 0{\cdot}3$	$t = 0{\cdot}632$	$P > 0{\cdot}5$	$t = 18{\cdot}060$	$P < 0{\cdot}001$
A *vs* D	$t = 0{\cdot}756$	$P < 0{\cdot}5$	$t = 4{\cdot}021$	$P > 0{\cdot}002$	$t = 5{\cdot}386$	$P < 0{\cdot}001$
A *vs* E	$t = 4{\cdot}301$	$P < 0{\cdot}002$	$t = 0{\cdot}200$	$P > 0{\cdot}8$	$t = 2{\cdot}895$	$P < 0{\cdot}005$
A *vs* F	$t = 4{\cdot}847$	$P < 0{\cdot}001$			$i = 1{\cdot}383$	$P < 0{\cdot}2$

to those already existing in mammals[11–12]. The results of this study indicate that the administration of homoplastic pituitary homogenates to induce ovulation leads to the production of steroids by both the ovaries and the adrenals. However, the adrenals seem to contribute a major share in the production of ovulatory steroids and the same might explain the facilitatory role played by the adrenocorticoids during ovulation. The physiological status of adrenals is, therefore, an important factor that should be taken into account for evaluating the ovulatory response of ovaries in Anura.

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1. Pesonen, S. and Rapola, J., *Gen. Comp. Endocr.*, 1962, **2**, 425.
2. Chieffi, G. and Botte, V., *Riv. Istoch. Norm. Pat.*, 1963, **9**, 172.
3. Botte, V. and Cottino, E., *Bull. Zool.*, 1964, **31**, 491.
4. Saidapur, S. K. and Nadkarni, V. B., *Curr. Sci.*, 1972, **41**, 531.
5. Ozon, R., Breuer, H. and Lisboa, B. P., *Gen. Comp. Endocr.*, 1964, **40**, 577.
6. Chang, C. Y. and Witschi, E., *Endocrinol.*, 1957, **61**, 514.
7. Ramaswami, L. S. and Lakshman, A. B., *Nature*, London, 1958, **182**, 122.
8. Werblin, H., Poltz, J., Le Roy G. V. and Davis, E. M., *J. Am. Chem. Soc.*, 1957, **79**, **10**.
9. Ramaswami, L. S. and Lakshman, A. B., *Proc. Nat. Inst. Sci. India*, 1959, **25**, 68.
10. Koche, F. C. and Hanke, M. E., *Practical Methods in Biochemistry* 220, Bailliere Tindal and Cox, London (1953). p, 220.
11. Callard, T. P. and Leathem, H., *Gen. Comp. Endocr.*, 1966, **7**, 80.
12. Redshaw, M. R. and Nicholls, J. J., *Ibid.*, 1971, **1**[illegible], 85.

A NEW TYPE OF EMBRYO SAC ORGANIZATION IN ANGIOSPERMS

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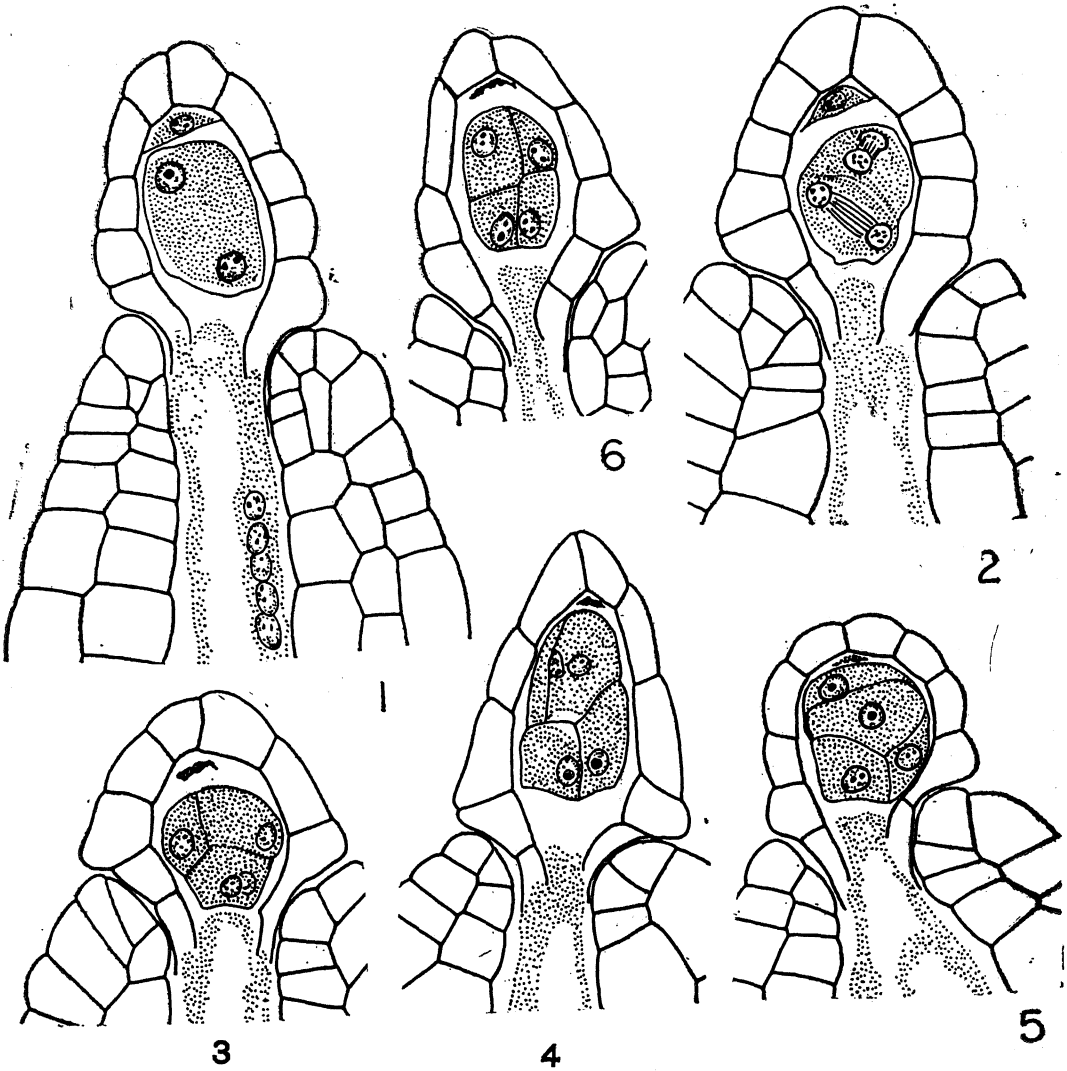
IN the course of our investigation on the development and organization of the embryo sac in Podostemaceae members, we have come across a peculiar type of organization so far not reported in any angiosperm.

The taxon *Willisia selaginoides* (Bedd.) Warming *ex* Willis was collected on 10-2-1973 in a stream flowing at Parambikulam submergible area in Trichur district, Kerala State. Voucher specimen bearing No. 63 *Nagendran* has been deposited in the herbarium of the botany department of the University of Mysore. FAA fixed flower buds were processed in alcohol-xylol series and serial longitudinal sections of 8–14 microns were cut using a 'Lipshaw' microtome. Staining was done in iron alum-hematoxylin, with erythrosin in clove oil as counterstain.

The bicarpellary syncarpous bilocular ovary has many anatropous tenuinucellate and bitegmic ovules, the outer integument alone organizing the micropyle. In each ovule, a hypodermal archesporial cell directly functions as the megaspore mother cell. After meiosis-I, it produces two dyad cells. The nucleus of the lower functional dyad cell completes meiosis-II. The two resulting megaspore nuclei are of equal size and a vacuole beween them is not organized (Fig. 1). Both the nuclei divide simultaneously (Fig. 2). Their spindles are disposed in a decussate manner. As a result. the four nuclei produced are correspondingly located within the embryo sac. When organization of the sac occurs, the two large synergids remain generally juxtaposed and are usually confined to the micropylar half of the ovoid embryo sac, while the egg and the



FIGS. 1–6. Development and organization of embryo sac in *Willisia selaginoides* (All Figs. × 900).
L.S. of an ovule showing 2-nucleate embryo sac and degenerated micropylar dyad cell; note organiza-
nucellar plasmodium. Fig. 2. Synchronous decussate division of the two nuclei in the embryo
3–5. Organized embryo sacs with two synergids, an egg and a polar cell in tetrahedral tetrad
egg and polar cell juxtaposed. Fig. 6. Four cells of organized embryo sac arranged in
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polar cell occupy the chalazal half of the sac, also juxtaposed (Figs. 3–6). The embryo sac, therefore, consists of four decussate cells although rarely an isobilateral arrangement is also noticed. Organization of the nucellar plasmodium will be complete when the ovule attains the 2-nucleate embryo sac stage.

In the hitherto investigated taxa of Podostemaceae the sequence of development and organization of the embryo sac conforms to the Apinagia or the Podostemum or the Polypleurum types, all of which are exclusive to this family[1–4]. The organized embryo sac of the Apinagia type has nuclear derivatives from the micropylar megaspore nucleus only, and consists of two synergids, an egg and a polar cell. This type of embryo sac has been recorded in the majority of the investigated taxa of the family[5]. In the Podostemum type, both chalazal and micropylar megaspore nuclei of the 2-nucleate embryo sac, contribute nuclei to the organized embryo sac also consisting of the usual egg apparatus and a polar cell[2]. The Polypleurum type develops like the Podostemum type but the organized embryo sac has reverse polarity. It consists of an egg apparatus at the chalazal end and a polar cell at the micropylar end; there are no antipodal cells[6].

The organized embryo sac of *Willisia selaginoides* has nuclear derivatives from both micropylar and chalazal nuclei of the 2-nucleate embryo sac and in this respect, it is similar to the Podostemum and the Polypleurum types in being truly bisporic. While the organized embryo sac of Polypleurum type presents an inverted image of the Podostemum type, the organization in *W. selaginoides* of the present study is totally distinct and the complements of the embryo sac are disposed in unusual planes. Since the organized embryo sac in *Willisia* is different from all other investigated angiosperms, it is appropriate to designate it as the " Willisia type ". The Willisia type is characterised by a bisporic, 4-nucleate embryo sac where the components are disposed in a decussate or isobilateral manner.

1. Battaglia, E., *Caryologia*, 1971, **24**, 401.
2. Arekal, G. D. and Nagendran, C. R., *Bot. Notiser* 1975 *a*, **128**, 332.
3. — and —, *Caryologiia*, 1975 *b*, **28**, 229.
4. Nagendran, C. R., Subramanyam, K. and Arekal, G. D., *Ann. Bot.*, 1976, **40**, 511.
5. —, *Curr. Sci.*, 1974, **43**, 259.
6. —, Arekal, G. D. and Subramanyam, K., *Amer. J. Bot.* (In press).

DIFFERENTIAL SUSCEPTIBILITY OF *ALLIUM CEPA* AND *A. TUBEROSUM* TO COLCHICINE TREATMENT

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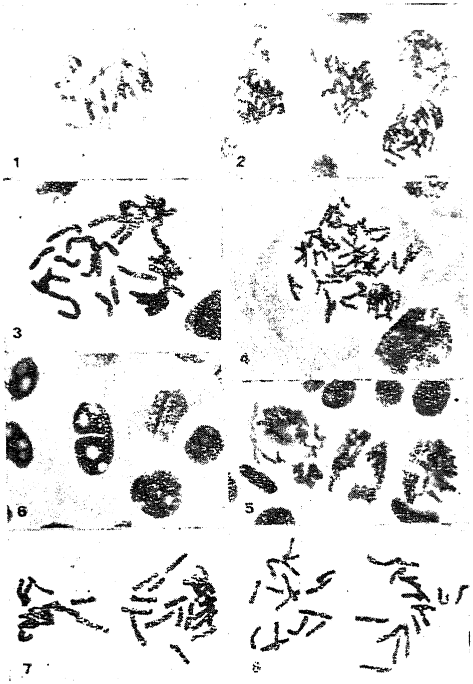
THE induction of polyploidy through colchicine in plant system is well established[1]. Inspite of its importance several species of plants fail to produce polyploids following colchicine treatment. There is either seedling lethality or reversion to diploid level after mixoploidy, through selective value of diploids at the initial stage. In such cases, low radiation after colchicine has been successfully applied in this laboratory[2] which allows the survival of polyploids with the elimination of diploids which are more radiosensitive.

As colchicine treatment is one of the important practices in improvements of crop species which are often met with failure, it was desired to determine precisely the degree to which polyploid and diploid species differ in their sensitivity to colchicine treatment, so that the data might help in programming of research on colchiploidy in plant system. Two allied species of *Allium*, viz., *A. cepa* and *A. tuberosum* were taken. *A. cepa* is an established diploid species ($2n = 16$), whereas *A. tuberosum* is an autotetraploid forming clear 8-quadrivalents in meiosis[3,4].

Young healthy bulbs and plants with fresh roots were taken and placed in jars containing the required concentration of colchicine for 24 h. at 20° C. After the treatment, the plants were taken out and placed for recovery in standard Knop's medium. After every 24 h., root tips were taken and fixed in acetic ethanol (1:2) for 2h. After rising in 45% acetic acid, root tips were heated in orcein mixture (2% acetoorcein: N·HCl: 9 : 1) for a few seconds, kept in that mixture for 1h and smeared in 45% acetic acid.

Of the different concentrations of colchicine tried 0·25% and 0·5% yielded very satisfactory results Cytological manifestation revealed marked differences. of colchicine effect between *A. cepa* and *A. tuberosum*. In *A. cepa*, clear spindle arrest was noted after colchicine treatment followed by the appearance of polyploid cells after recovery (Figs. 1, 2).

In *A. tuberosum* on the other hand, appearance of cells with chromatids often separated as in anaphase but with no poles indicating arrest, was found to be present in approx. 20% of the dividing cells. Here individual members represent chromatids instead of chromosoms (Fig. 4). But no clear polyploid cell was observed, excepting in one case out of 200 dividing nuclei studied. Moreover, the formation of chromosome groups even with nuclear membrane intact has been recorded (Fig. 5). The total number of chromosomes involved in the grouping did not exceed 32. In metabolic nuclei too, occasional tendency of cleaving into two groups was observed (Fig. 6). Another feature was the separation of chromosomes as such, with two chromatids intact to two sides of the nucleus showing reductional groupings in approximately 50% of the cells. Of these again, such reduction may be of unequal number of chromosomes in two poles (18–20%), but in most cases (70–80%) separation involved 16 chromosomes (Figs. 7, 8).



FIGS. 1–8. Colchicine action on *Allium cepa* and *A. tuberosum*. Figs. 1–2. Diploidy and induced tetraploidy in *A. cepa*. Figs. 3–8. *A. tuberosum*. Figs. 3–4. Natural tetraploid and diplochromatid appearance following treatment. Fig. 5. Chromosome grouping. Fig. 6. Tendency of nuclear cleaving at the metabolic stage. Figs. 7–8. Somatic reduction showing unequal and equal separation respectively.

The absence of polyploidy through colchicine treatment even after spindle disturbance and chromatid separation without pole formation indicates that at this autotetraploid species there is an inbuilt genetic mechanism to check uninterrupted increase of chromosomes through duplication. It is operating through no further division of cells thus affected after colchicine treatment, resulting in almost absence of polyploid cells after recovery.

The formation of groupings hinder the origin of polyploid cells. A reflection of the inherent mechanism of checking polyploidy is the reductional separation of chromosomes. As each chromosome in diploid nuclei contains two chromatids, there is the possibility of restoration of tetraploid level in the next cell cycle. In otherwise apparently autotetraploid species where cryptic gene changes might have been involved in some of the chromosomes not sufficient to hinder multivalent formation, such reductional separation followed by duplication may prove to be helpful in securing a true autotetraploid level in relation to such cryptic alterations. Such alterations may include some controlling units as well.

The above data may be taken to indicate that in this autotetraploid species, there is an inbuilt mechanism of resisting colchicine action. Moreover colchicine treatment in such species may be utilized to induce somatic reduction. Nuclei with reduced chromosomes may help on the one hand the recovery of diploids and on the other, if they form tetraploids as well, the homozygosity of certain gene changes may be secured at tho tetraploid level. Investigations are in progress to find out the applicability of this behaviour in other species as well.

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1. Kihlman, B. A., *Action of Chemicals on Dividing Cells*. Prentice-Hall, Inc., Englewood, Cliffs, New Jersey, 1966.
2. Das, A., *Ph.D. Thesis.*, Department of Botany, University of Calcutta, 1972.
3. Kurita, M., *Mem. Ehimc Sec. II, Ser.* B4, 1962, p. 403.
4. Sen, S., *Caryologia*, 1974, **27**, 7.

LETTERS TO THE EDITOR

APPLICATION OF Cu (II)-THIOMALLIC ACID COMPLEX AS AN INDICATOR IN THE COMPLEXOMETRIC DETERMINATION OF Al (III), Ga (III) AND In (III)

SEN SARMA[1] has reported the use of thiomallic acid and Fe (II)-thiomallic acid as an indicator in the estimation of Fe (II), Cu (II), Mn (II) and Ca (II), Mg (II) and Pb (II) respectively. Since then, little effort has been put, to utilize the comparatively cheaper indicator to estimate other metal ions. Here is an attempt towards the estimation of Al (III), Ga (III) and In (III) using Cu-thiomallic acid complex as an indicator.

Cu (II) forms a violet complex with thiomallic acid (TMA) in the pH range 4·8 and 5·0. The complex is stable up to 40° C, above which it decomposes. The aliquots of the solution containing 7 to 20 mg of In (III), when titrated against a standard EDTA solution in the presence of Cu (II)-TMA complex, gives a sharp colour change from violet to greenish blue at the end point. In the case of Al (III) and Ga (III) the method of back titration was employed when the colour change was from blue to violet at the end point. The results, when compared with those obtained by the standard methods described in the literature[2], are found to be nearly the same (Table I).

TABLE I

Metal ion	Amount of metal ions in mg.	
	Taken	Found (mean of 4 values)
Al (III)	(1) 4·58	4·583
	(2) 11·14	11·145
	(3) 20·27	20·265
Ga (III)	(1) 8·40	8·45
	(2) 17·81	17·75
	(3) 28·24	28·10
In (III)	(1) 6·84	6·85
	(2) 12·44	12·46
	(3) 21·10	21·04

The ions like Pb (II), Cd (II), Zn (II), Fe (II), Co (II), Th (IV), CNS^-, I^-, CrO_4^{2-} interfere seriously and should be absent. The special feature of the proposed method is the use of a relatively cheaper reagent (TMA) and the sharp end point which helps in the accurate determination of the cations under study.

Experimental

Solutions of Al (III), Ga (III) and In (III) were standardized by the usual methods[2].

Reagent.—One per cent solution of thiomallic acid was prepared in double distilled water.

Estimation of In (III)

An aliquot of the solution containing 7 to 20 mg of In (III) is mixed with 1 ml of the standardized Cu (II) solution and 1 ml of TMA. The pH of the mixture is adjusted between 4·8 and 5 using sodium acetate acetic acid buffer. The mixture is titrated with a standard solution of EDTA until the colour changes from violet to greenish blue. The amount of EDTA required for 1 ml of Cu (II) solution being known, the amount of In (III) in the aliquot can be calculated.

Estimation of Al (III) and Ga (III)

An aliquot of the solution containing 5 to 30 mg of Al (III) or Ga (III) is added to a known excess of EDTA and 10 ml of buffer solution (pH 4·8 to 5). The mixture is heated to near boiling and cooled. The pH of the mixture is then adjusted between 4·8 and 5 as above. The TMA solution (1 ml) is added and the mixture is titrated against the standard solution of Cu (II) until the colour changes from blue to violet. The amount of EDTA initially added being known, that consumed by Al (III) and Ga (III) can be calculated.

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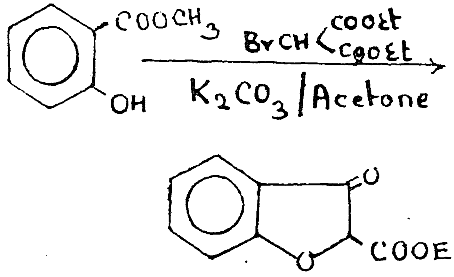
1. Sen Sarma, R. N., *Sci. and Cult.*, 1958, 23 (8), 434.
2. Vogel, A. I., *Quantitative Inorganic Analysis*, III Edn., Longmans Green and Co. Ltd.

A CONVENIENT METHOD FOR THE SYNTHESIS OF 2-CARBETHOXY 3 (2 H) BENZOFURANONE

SCHRODER *et al.*[1], have made an investigation of a number of available methods for the synthesis of 3 (2 H) benzofuranone and found that Dieckmann cyclisation of the ethyl 0-carbethoxymethyl salicylate was the most effective method. This requires the initial preparation of 0-carbethoxymethyl salicylate by the condensation of ethyl chloroacetate or ethyl bromoacetate with *n*-ethyl salicylate. We felt that both condensation and cyclisation may be effected in a single stage by the use of ethyl bromomalonate instead of ethyl bromoacetate. Because it is shown that the condensation of ethyl bromomalonate with salicylaldehyde in presence of anhydrous potassium carbonate in dry acetone resulted in the simultaneous condensation and cyclisation accompanied with the partial hydrolysis and decarboxylation giving benzofuran 2-carboxylate[2], in a single step. It is also reported that the condensation of chloroacetone with methyl salicylate in the presence of a large excess of anhydrous potassium carbonate gave some amount of cyclised product, *viz.*, 2-acetyl-3 (2 H) benzofuranone[3]. As we needed 2-carbethoxy-3 (2 H) benzofuranone in connection with our work, we have made an attempt to obtain it in a single step by condensing ethyl bromomalonate with methyl salicylate in presence of anhydrous potassium carbonate. As expected condensation, cyclisation. partial hydrolysis and decarboxylation occurred simultaneously and 2-carbethoxy 3 (2 H) benzofuranone was obtained 60% yield. The identity of the compound was established by its m.p. and superimposable ir and uv spectra with that of an authentic sample obtained by known method[4].



Experimental

2-Carbethoxy-3 (2 H) benzofuranone.—A mixture of freshly distilled methyl salicylate (30·4 g), ethyl bromomalonate (48 g) and anhydrous potassium carbonate (60 g) in dry acetone (150 ml) was heated under reflux on a water bath while stirring magnetically for 12 hours. The reaction product was filtered and potassium salts were washed with dry ether until colourless. The dry salt was suspended in water (200 ml) and cooled thoroughly in ice bath. The suspension was carefully acidified with dilute acid and the 2-carbethoxy-3 (2 H) benzofuranone which separated as a colourless solid was collected by filtration. On crystallisation from petroleum ether it was obtained as colourless needles. m.p. 63–64° (lit. m.p. 65°), yield, 25 g. (Found C, 63·81 ; H, 4·57 ; $C_{11}H_{10}O_4$ requires C, 64·08 ; H, 4·85%.)

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---

1. Schroeder, D. C., Corcoran, P. O., Holden, C. A. and Mulligen, M. C., *J. Org. Chem.*, 1962, 27, 586.
2. Kawase, Y., Nakayama, M. and Tamatskuri, P., *Bull. Chem. Soc.*. Japan, 1962, 35, 149.
3. Geissman, T. A. and Armen, A., *J. Am. Chem. Soc.*, 1955, 77, 1623.
4. King, A. I., Holland. R. A., Reed, F. P. and Robertson, A., *J. Chem. Soc.*, 1948, p. 1672.

SPECTROPHOTOMETRIC STUDIES ON Pd (II)-5-CHLORO-2-HYDROXY-4-METHYL ACETOPHENONE OXIME COMPLEX

5-CHLORO-2-hydroxy-4-methylacetophenone oxime (CHMAO) is a good complexing agent for a number of transition metal ions[1]. The literature reveals that the spectrophotometric studies of its palladium chelate have not been carried out. In continuation[2,3] of our work on disubstituted *o*-hydroxy acetophenone oximes, we now report the spectrophotometric studies of the palladium complex of CHMAO by its extraction into chloroform.

Experimental

Apparatus.—Optical density and pH measurements were carried out with the Systronic, Colorimeter type (103) and type (322) respectively.

Solution.—Standard solution of palladium (II) was prepared from palladium chloride, B.D.H. (A.R.) in dilute hydrochloric acid and standerdized gravimetrically[4]. Other solutions were prepared from AnalaR reagents. Hydrochloric acid and ammonium hydroxide buffers of different pH values were used.

The reagent (CHMAO) was prepared as reported earlier[1]. Freshly distilled chloroform (A.R. grade) was used for the extraction of the complex.

Procedure for the determination of palladium.—A suitable aliquot of palladium (II) solution

[(10–40 μg Pd (II)] is mixed with CHMAO (2 ml., 0·01 M ethanolic solution). The mixture was buffered to pH 2–4 and the total volume made upto 10 ml with water. The mixture was allowed to stand for about 100 min. and then extracted into 8 ml. of chloroform by vigorous shaking. The chloroform solution was separated and the absorbance recorded against the reagent blank prepared under similar conditions at 400 nm. The palladium content was found from the calibration curve.

Interferences.—The following ions did not cause a deviation in absorbance of more than 2·5% at pH 2·5–4·0 when present in amounts (ppm) shown in parentheses: Co (II) (500), Ni (II) (500), Cu (II) (300), Zn (II) (800), Mg (II) (800); Cd (II) (1000), MoO_2 (II) (500), UO_2 (II) (500), VO (II) (500), Hg (II) (500), Mn (II) (500), Sb (III) (400), Bi (III) (400), As (III) (400), Cl^-, Br^-, I^-, NO_2^-, $C_2O_4^-$, SO_4^{2-}, SO_3^{2-} and CO_3^{2-} upto (2000 each). But EDTA, Fe (III), and F^- interfere seriously even in small quantities.

Results and Discussion

Palladium forms yellow precipitate with CHMAO, which is extractable into chloroform easily. Vosburgh and Cooper's method[5] shows the formation of only one complex and maximum absorbance lies at 400 nm. The absorbance *versus* pH plot shows that the absorption of the complex remains maximum and constant between pH 2·5–4·0 and pH 3·0 was maintained for these studies. Similarly absorbance *versus* reagent plot shows that at least four times excess of reagent is essential for complete precipitation.

The system obeys Beer's law upto 18 ppm of palladium and from Ringbom's plot is 5·00–16·20 ppm. The molar extinction coefficient is 2800 and its sensitivity is 0·038 μg Pd/cm^2.

Stoichiometry of the complex.—Job's method[6], mole ratio method[7] and slope ratio method[8] showed that the metal and the ligand were present in 1 : 2 ratio in the complex.

Stability constant of the complex.—The conditional stability constant (27° C) was determined by (1) the mole ratio method and (2) Dey and Mukherji method[9] and the log of stability constant was 8·44 and 8·76 respectively. The free energy of formation of the complex, $- \Delta F = RT \ln K$, was 11·86 Kcal/mole.

A number of oximes react with palladium (II) to give precipitate which are extractable into organic solvent like chloroform. Dimethylglyoxime[10] is a selective reagent for separation of palladium (II) from other platinum metals. But the sensitivity is poor (0·06 μg Pd/cm^2) at 375 nm. α-Furildioxime[11] (sensitivity 0·005 μg Pd/cm^2 at 380 nm) and phenyl-α-pyridyl ketoxime[12] (sensitivity 0·0036 μg Pd/cm^2 at 410 nm) are sensitive and selective reagents for palladium (II). However, CHMAO (sensitivity 0·038 μg Pd/cm^2 at 400 nm) is more sensitive than dimethylglyoxime but less sensitive and selective as compared with the other oximes.

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1. Saksena, R. N. and Panday, K. K., *J. Ind. Chem. Soc.*, 1972, 49, 779.
2. Gupta, S. P., Srivastava, S. K. and Lal, K., *Ind. J. Chem.*, 1975, 13, 297.
3. Lal, K. and Gupta, S. P., *Curr. Sci.*, 1975, 44, 178; 652. *Ind. J. Chem.*, 1975, 13, 973 (in press).
4. Vogel, A. I., *A Text Book of Quantitative Inorganic Analysis*, Longmans Green and Co., Ltd., London, 1962, p. 511.
5. Vosburgh, W. C. and Cooper, G. R., *J. Am. Chem. Soc.*, 1941, 63, 437.
6. Job, P., *Ann. Chim.*, 1928, 9, 113.
7. Goe, J. H. and Jones, A. L., *Ind. Eng. Chem., Anal. Ed.*, 1944, 16, 111.
8. Harvey, A. E. and Manning, D. L., *J. Am. Chem. Soc.*, 1950, 72, 4488.
9. Mukherji, A. K. and Dey, A. K., *J. Inorg. Nucl. Chem.*, 1958, 6, 314.
10. Peshkova, V. M., Shlenskaya, V. I. and Rashevskaya, A. I., *Anal. Abs.*, 1957, 4, 2623.
11. Menis, O. and Rains, T. C., *Anal. Chem.*, 1955, 27, 1932.
12. Sen, B., *Ibid.*, 1959, 31, 881.

PREPARATION MOLAR CONDUCTANCE AND INFRA RED SPECTRA OF SOME AROMATIC AMINE COMPLEXES OF COPPER (II) PHTHALIMIDE

Part I. Complexes of *o*–, *m*–, *p*–toluidines, piperidine and morpholine

THE above-mentioned amines have been used to prepare 5 new coordination complexes with copper (II) phthalimide, and their structures have been resolved on the basis of analysis, molar conductance and infra red measurements.

All the complexes were prepared by refluxing copper (II) phthalimide with amines in 1 : 2 ratio in acetone for 6 hours, and crystallising out the complex from solution by the addition of petroleum ether. These were recrystallised from methanol, dried and analysed.

(*a*) *Diphthalimidodipiperidine Copper (II)*—$[Cu(C_8H_4O_2N)_2\,pip_2]^\circ$. Found Cu = 13·90; C = 62·97; H = 5·89; N = 10·98%; Calculated Cu = 14·25; C = 63·34; H = 6·09; N = 11·34%.

TABLE I

Infrared absorption frequencies in Cm^{-1}

Complex	molar conductance	NH stretching	NH$_2$ deformation	C–N stretching	H–C–C– wagging	H–N–C deformation	>C=O stretching
(*a*) $[Cu(C_8H_4O_2N)_2(pip)_2]^\circ$	0·37	3160 s	..	..	1470 s 1380 m 1300 m	850 m	1683 s
(*b*) $[Cu(C_8H_4O_2N)_2(mor)_2]^\circ$	0·32	3210 s	..	..	1420 s 1370 m 1335 m	858 w	1691 s
(*c*) $[Cu(C_8H_4O_2N)_2(o\text{–}tol)_2]^\circ$	0·33	3250 s	1600 s	1110 s	..	..	1680 vs
(*d*) $[Cu(C_8H_4O_2N)_2(m\text{–}tol)_2]^\circ$	0·31	3220 s	1609 m	1109 s	..	..	1692 s
(*e*) $[Cu(C_8H_4O_2N)_2(p\text{–}tol_2]^\circ$	0·39	3270 m	1601 s	1095 s	..	..	1688 s

(*b*) *Diphthalimidodimorpholine Copper (II)*—$[Cu(C_8H_4O_2N)_2 mor_2]^\circ$ Found Cu = 12·57 ; C = 58·08 ; H = 4·88 ; N = 10·90% ; Calculated Cu = 12·76 ; C = 57·88 ; H = 5·22 ; N = 11·25%.

(*c*) *Diphthalimidodi* (*o-toluidine*) *Copper* (*II*)—$[Cu(C_8H_4O_2N)(o\text{-}tol)_2]^\circ$ Found Cu = 11·55 ; C = 66·62 ; H = 4·61 ; N = 9·96% ; Calculated Cu = 11·81 ; C = 66·97 ; H = 4·82 ; N = 10·41%.

(*d*) *Diphthalimidodi*(*m-toluidine*) *Copper* (II)—$[Cu(C_8H_4O_2N)_2(m\text{-}tol)_2]^\circ$ Found Cu = 11·67 ; C = 66·89 ; H = 4·51 ; N = 9·82%.

(*e*) *Diphthalimidodi* (*p-toluidine*) *Copper* (*II*)—$[Cu(C_8H_4O_2N)_2(p\text{-}tol)_2]^\circ$. Found Cu = 11·98 ; C = 66·66 ; H = 4·92 and N = 10·02%.

Copper was estimated as cuprous thiocyanate, Carbon, hydrogen and nitrogen were estimated microanalytically. Conductivity values were determined in methanol solution (Conc. 10^{-3} M) with Philips PR 9500/90 instrument. I.R. spectra were recorded with nujol mulls on Perkin Elmer Spectrophotometer model 137. Details are given in Table I.

The molar conductance values for the complexes are in the range 0·3–0·6 and these confirm their non-electrolytic nature. It is thus indicated that both the amine and phthalimide ions are coordinated to the metal and the number of amine molecules coordinated is 2 for all the amines used.

In the i.r. spectra of the toluidine complexes the N–H stretching, NH$_2$ deformation and C–N stretching vibrations show considerable negative shift which has been taken as an evidence for coordination through the nitrogen atom of the base[1-5]. In the piperidine and morpholine complexes, the bands due to NH stretching vibrations show negative shift, but those due to H–C–C wagging and H–N–C deformation modes show positive shift, due to the coordination of the nitrogen atom of the amine[6].

All complexes show one strong band at around 1685 ± 10 cm^{-1} which is assigned to be the $>C=O$ stretching vibration, which shows a negative shift of the order of some 60 cm^{-1} when compared to free phthalimide. This is again taken as an evidence of the coordination of nitrogen of the phthalimide ion[7-8].

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1. Fowles, G. W. A., *Proceeding Symposium Chem. Coordination Compounds,* Pt. 2, p. 41.
2. Rao, C. N. R. and Venkatraghvan, R., *Can. J. Chem.*, 1961, 39, 1757.
3. Misra, C. H., Parmar, S. S. and Shukla, S. N., *J. Inorg. Nucl. Chem.*, 1966, 28, 147.
4. Ahuja, I. S. and Rastogi, P., *Ibid.*, 1970, 32, 2085.
5. Rao, C. N. R., *Chemical Application of Infra Red Spectroscopy.* Academic Press, New York, 1963, p. 250.
6. Greenwood, N. N. and Wade, K., *J. Chem. Soc., A.* 1960, p. 1130.
7. Bagnoll, W., Deane, M., Robinson, S. and Stewart, A., *Ibid.*, 1961, p. 1611.
8. Jain, S. C. and Rivest, R., *Can. J. Chem.*, 1962, 40, 2243.

RHIZOBIUM INOCULATION AND OIL CONTENT OF SOYBEAN SEEDS *GLYCINE MAX* (L.) MERRILL

IT is reported that the oil per cent of soybean seeds decreases with an increase in yield, resulting from *Rhizobium* inoculation[1-3]. The results of two independent experiments with the strains of *R. japonicum* and their composites, presented in this paper, show that though, initially the oil per cent decreases with an increase in yield, at much higher yields, it becomes nearly constant. Field experiments were carried out at Bangalore[4] using soybean (*Glycine max*) var. *Davis*, by raising plants after inoculating seeds by the usual procedure. The oil per cent of the samples was determined by the pulsed NMR technique[5].

The results presented in Fig. 1 show that the oil per cent of the seeds decreases with increase in yield of grains and this in turn is related to the efficiency of *R. japonicum* strains. A quantitative examination of Fig. 1 *a* gives a value of 1·42 for the ratio of the highest yield to the control yield and the corresponding value for oil ratio is 0·81. A similar examination of Fig. 1 *b* gives a much higher value for the yield ratio (5·49) whereas, the corresponding oil ratio (0·84) is not lowered. This means that initially the oil per cent decreases significantly with the increase in yield over the control, at much higher yields the oil per cent becomes nearly constant at a reduced level and does not decrease further with increase in the yield.

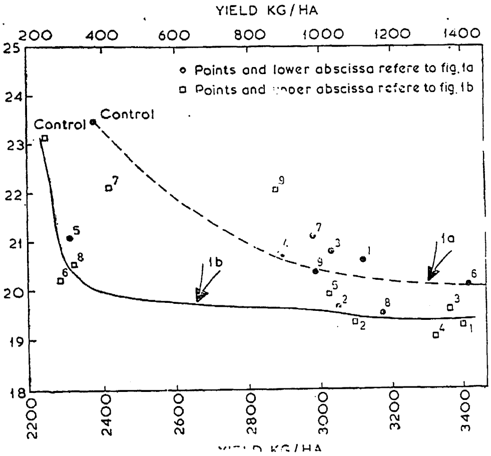

FIG. 1 *a–b*. Fig. 1 *a*. Oil content of soybean (var. *Davis*) in relation to yield due to inoculation with the following composites: (1) 'Nitragin', a mixed culture; (2) E 177 + E 188 + SB 6 + SB 16; (3) E 177 + E 188 + SB 6; (4) E 177 + SB 6; (5) E 177 + 188; (6) E 177 + SB 16; (7) E 188 + SB 6; (8) SB 6 + SB 16 and (9) SB 6 + SB 16 + USDA — 110. Fig. 1 *b*. Oil content of soybean (var. *Davis*) in relation to yield due to inoculation with different strains of *R. japonicum*: (1) USDA 10; (2) SB 6 (I.A.R.I.); (3) SB 16 (I.A.R.I.); (4) UPAU 3; (5) Kanpur cult.; (6) E 177–(J.N.K.V.V.); (7) E 188–(J.N.K.V.V.); (8) P 19 (Hissar) and (9) S 5 (Bose Institute, Calcutta).

Table I provides data on oil production (oil per cent × yield) per hectare, which reveals the fact that in spite of the observed decrease in oil per cent due to *Rhizobium* inoculation, the net oil production per hectare in the inoculated series is far greater than uninoculated series. This is because the increase in yield due to the inoculation is much more than the decrease in the oil per cent.

TABLE I

Influence of Rhizobium *inoculation on the oil content of soybean seeds* (*var.* Davis)

Treatment	Oil production kg/ha* (oil per cent × yield)
Control plants (Uninoculated)	54·12
Inoculated plants	
Hissar (P 10)	77·40
E 177	92·06
E 188	98·79
Bose Insttitute (S 5)	202·58
Kanpur cult.	207·06
SB–6	208·22
UPAU 3	248·32
USDA 110	266·70
SB 16	268–41

* Results are the average of four replicates.

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1. George, A. H. and Erdman, W., *Agron. J.*, 1964, 56, 423.
2. Weber, C. R., *Ibid.*, 1966, 58, 43.
3. Caldwell, B. E., *Ibid.*, 1969, 61, 813.
4. Balasundaram, V. R., Report from I.A.R.I., Centre-Microbiological Experiments conducted Soybean during *kharif*, 1972, data presented at VI Soybean Workshop, held at Dharwar (Mysore State), 1973.
5. Tiwari, P. N., Gambhir, P. N. and Rajan, T. S., "Rapid and non-destructive determination of seed oil by pulsed NMR technique," *J. American Oil Chemist's Soc.*, 1974, 51, 104.

SENSE ORGANS ON THE PHARYNGEAL PLATE OF THE ANT *CAMPONOTUS COMPRESSUS* FABR. (HYMENOPTERA: FORMICIDAE)

THE pharyngeal sense organs in ants have so far been reported only in *Myrmica rubra*[1]. These sense organs occur not only in Formicoidea but also in other Hymenoptera including the honey-bee[2]. The present author has found them also in certain wasps, ichneumonids and sphecoids. Among the ants, she has found them in *Cataglyphis setipes* Forel and *Camponotus compressus* Fabr. (Formicinae), *Messor barbarus* Linn. (Myrmicinae), *Dorylus orientalis* Westwood and *Brachyponera* sp. (Dorylinae), *Detraponera rufonigra* Jerdon (Pseudomyrmicinae). These sense organs have the same general pattern in all these ants. The present study deals with the structure and significance of these sense organs in *Camponotus compressus* Fabr. and is based workers, soldiers and alates (males and females) of *C. compressus* collected locally from Delhi.

The pharyngeal sense organs are located on a ventral plate in the anterior part of the pharynx (Fig. 1). The ventral plate (Fig. 2) is highly sclerotised at its lateral margins, the rest of it is comparatively thin and is further differentiated into two lateral areas each of which bears 30–45 microscopic transparent oval discs (*s.p.*), which measure about 11 × 7·5 microns. In stained microscopic preparations, these oval discs are found to be the external terminations (sense plates of the sensilla placodea (*sn. pl.*).

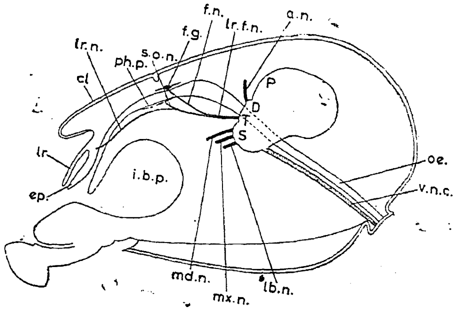

FIG. 1. Diagrammatic sketch to show the location and the nerve supply of the sense organs of pharynx of *Camponotus compressus*.

The microtome sections of the ventral plate show that the external sense plate is a thinned out portion of the cuticle which overlies a minute oval hollow gap (Fig. 3 *b*). Such areas, in the surface view, appear as transparent discs because of their thinness (about 1 micron).

Each sense plate bears internally one sensillum placodeum consisting of the usual envelope cell (*e.c.*), a cap cell (*c.c.*) and a set of 4 to 6 bipolar sense cells (*s.c.*) (Fig. 3 *a*)[3]. The pharyngeal sense organs are innervated from a nerve arising from the labral branch of the principal labro-frontal nerve (Fig. 1, *lr.f.n.*).

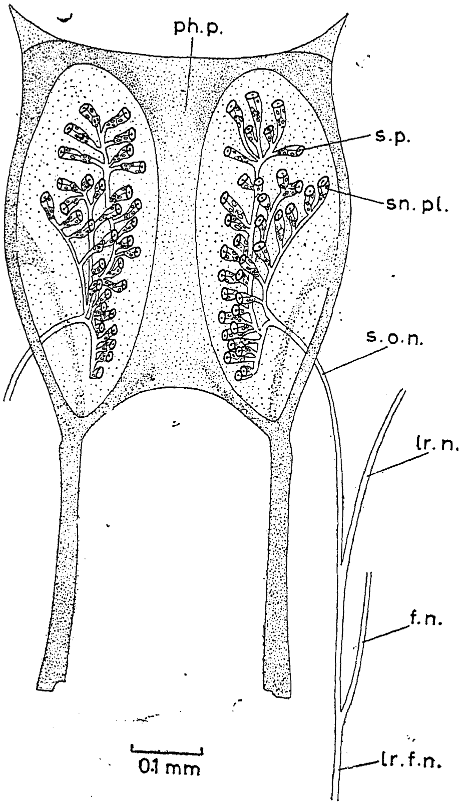

FIG. 2. Pharyngeal plate with the attached underlying sense organs.

The sensilla placodea occur on the antennae of certain Hymenoptera, Homoptera and Coleoptera[3]. These sensilla on the antennae of honey-bee were experimentally confirmed to be olfactory receptors[4,5]. However, the mode of perception of odour stimuli through the thick sclerotised sense plates (approximately 1·5 microns) in honey-bee is not yet understood[3,6,7].

The sensilla placodea here described in *Camponotus compressus* have not been hitherto recorded. They are undoutedly sensilla placodea because of their characteristic histological structure comprising of a sense plate, an envelope cell, a cap cell and a group of sense cells (Fig. 3). Their location on the pharyngeal plate suggests that these are gustatory or contact chemoreceptors, because the food ingested would directly come in contact with them. The gustatory receptors hitherto reported in other insects are sensilla trichodea and sensilla basiconica, as in some Diptera, Lepidoptera, Hymenoptera and Coleoptera[7].

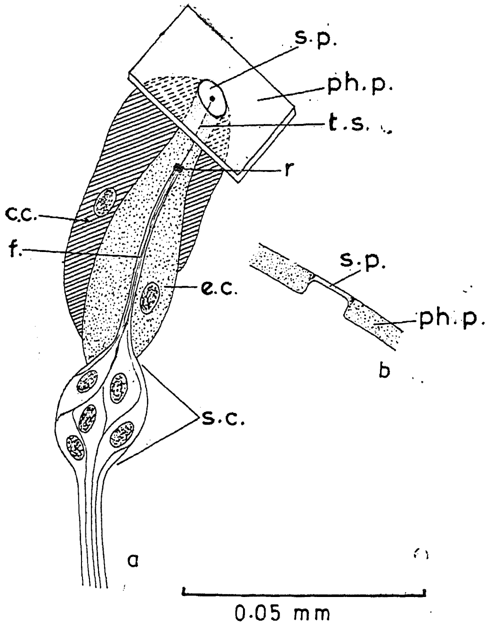

FIG. 3 *a, b.* Diagrammatic sketch of sensillum placodeum and the sense plate.

The pharyngeal sense organs are present in most hymenopterans, but in ants these appear to be more deep-seated. In ants, the food is first received in the infra-buccal pouch (Fig. 1, *i.b.p.*) and then passes to the pharynx *via* a long food meatus. Other hymenopterans have a relatively shorter food meatus. The pharyngeal sense organs in those cases might serve only for taste. However, the deeper location of pharyngeal sense organs in *C. compressus* suggests that these may have an additional role to play. In ants, the food collected by a worker is shared with the other nestmates including males, females and also the larvae. The regurgitative feeding behaviour in adult ants can be initiated or terminated by either the donor or the acceptor. The antennae and forelegs are used in the correct orientation of the ants and in regurgitation[8].

It is known that the workers lick up secretions of larvae from the body which are apodous and lack antennae. It is believed that the act of licking stimulates the workers to offer food to the larvae. At this stage it is not possible to identify the pathway which results in disgorging of the food by the workers, although a connection exists between the sense organs and the sympathetic nervous system *via* labral nerve and the frontal ganglion.

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---

1. Janet, C., "Observations sur les fourmis," *Limoges, Ducourtieux et Gout,* 1904; p. 68.
2. Snodgrass, R. E., *Anatomy and Physiology of Honey-bee,* Comstock Publ., New York, 1956.
3. —, *Principles of Insect Morphology,* McGraw-Hill, New York, 1935.
4. Von Frisch, K., *Zool. Jhrb. Zool. Physiol.,* 1921, p. 449.
5. Dostal, B., *Zeit. Vergl. Physiol.,* 1958, **41,** 179.
6. Slifer, E. H., *International Review of Cytology,* 1961, 2, 125.
7. Dethier, V. G., *The Physiology of Insect Senses,* Chapman and Hall Science Paperbacks, 1963.
8. Wallis, D. I., *Behaviour,* 1961, **17** (1); 17.

NEUROCHEMICAL CORRELATES OF ALLOXAN-DIABETES: CHANGES IN THE AMINOTRANSFERASE ACTIVITY IN THE BRAIN STEM REGION OF FROG

INFORMATION concerning the changes in enzyme activities during alloxan-diabetes in various tissues of mammals is vast[1-6]. However, very little information is available on the activities of various enzymes in amphibians during alloxan-diabetes[7,8]. Since the brain stem showed profound changes compared to other regions of the brain in certain biochemical parameters[9,10,11] (like levels of protein, glycogen and RNA) during the course of our present investigation on neurochemical correlates of alloxan-diabetes, it was felt desirable to initiate the present study. Since alloxan-diabetes induces significant changes in protein[11] and RNA[11] levels in the frog, *Rana cyanophlictis,* it is likely that aminotransferases, which are known to link amino acid and carbohydrate metabolism[12], should also be affected. Hence, in the present investigation, the activity levels of Aspartate aminotransferase (AAT, E.C. 2.6.1.1) and Alanine aminotransferase (AlAT, E.C. 2.6.1.2) were studied in the brain stem of frog, on inducing alloxan-diabetes.

Frogs, *Rana cyanophlictis,* of medium size (22–26 g) were maintained in the laboratory (25 ± 2° C) in glass aquaria in tap water. Diabetes was induced by intramuscular injection of alloxan monohydrate as described earlier[11].

TABLE I

Aminotransferase activities in proportion to enzyme concentration in the brain stem of normal and diabetic frogs

Enzyme Concn. in mg	AAT		AlAT	
	Control	Experimental	Control	Experimental
2	0·11 ±0·002	0·14±0·016	0·135±0·03	0·155±0·015
4	0·15 ±0·003	0·19±0·005	0·17 ±0·001	0·185±0·02
6	0·175±0·01	0·23±0·02	0·21 ±0·04	0·23 ±0·06
8	0·20 ±0·025	0·26±0·04	0·255±0·007	0·27 ±0·05
10	0·24 ±0·006	0·31±0·01	0·28 ±0·01	0·315±0·08

No. of observations 5 (Mean ± S.D.).
Values are expressed as micromoles of Ketoacid formed per mg tissue per hour.
Values are significant; $P > 0·05$.

Frogs were decapitated and the brain was removed from the ventral side. The brain stem was separated using a sterilized scalpel by keeping the brain immersed in ice-cold frog's Ringer[13]. The tissues were weighed with Ringer in an electric balance and immediately used for analysis.

The activity levels of the aminotransferases were measured following the colorimetric procedure of Reitman and Frankel as described by Bergmeyer[14]. Blood glucose was estimated by the method of Folin[15].

The blood glucose level (54·6 ± 4·8 mg/100 ml) in 48 h alloxan-diabetic frog was 73% higher than in controls (31·5 ± 2·3 mg/100 ml). It is clear from Table I that the activity level of AlAT is higher than that of AAT in both normal and diabetic conditions. Further, marked elevation in the activity levels of AAT and AlAT in the brain stem of frog on inducing diabetes was noted (Table I).

The aminotransferase activities in various tissues are known to increase under several pathological and altered physiological conditions[12,16,17]. The associated physiological conditions are enhanced gluconeogenesis, increased protein catabolism and a rise in the amino acid pool[18].

An augmentation in the activity levels of aminotransferases observed in the present study during diabetes may be due to an increase in the free amino acid pools of the different regions of the brain of frog, *R. cyanophlictis* on inducing diabetes (Nayeemunnisa and Nagaraj, unpublished observations). It is of interest in this connection to note that the glutamic acid level exhibited dramatic increase (percentage change; 100–120% increase) over controls in the different regions of the brain of frog and rat during diabetes (Jayashree and Nayeemunnisa, unpublished data). It is, therefore, possible that the observed increase in the activity levels of AAT and AlAT is due to such a change occurring in the level of brain glutamic acid during diabetes.

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---

1. Shigeru, K., Hosoki, K. and Yoshimura, Y., *J. Biochem.* (Tokyo), 1971, 69 (1), 247.
2. Sarkisyan, L. V. and Adunts, G. T., *Biol. Zh. Arm*, 1969, 22 (10), 26.
3. Kittong and Chung, *J. Cath. Med. Coll.*, 1970, 15, 251.
4. Jakobsson, S. V. and Gustar Dallner, *Biochem. Biophys. Acta*, 1968, 165 (3), 380.
5. Sanford, M. and Newell, N. F., *Invest. Ophthalmol.*, 1971, 10 (6), 453.
6. Jayashree, C. and Nayeemunnisa, *Life Sci.*, 1975, 17, 1159.
7. Nayeemunnisa, and Jayashree, C., *Ind. J. Biochem. Biophys.*, 1976, Communicated.
8. — and Abdul Azeem, *Experientia,* 1976, Communicated.
9. Jayashree, C. and Nayeemunnisa, *Proceedings of the All India Symposium on Comparative Animal Physiology*, 1975.
10. — and —, *Experientia*, 1976, Communicated.
11. Nayeemunnisa, *Ibid.*, 1975, 31 (8), 942.
12. Harper, H. A., *Review of Physiological Chemistry* (Maruzen Asian Edition), 1965.
13. Lockwood, A. P. M., *Animal Body Fluids and Their Regulation*, 1963.
14. Bergmeyer, H. U., *Methods of Enzymatic Analysis,* Academic Press, New York, 1965, p. 837.
15. Oser, B. L., *Hawk's Physiological Chemistry*, (TMH Edition), 1965.
16. Nichol, C. A. and Rosen, F., *Weber Advances in Enzyme Regulation*, (Pergamon Press, N.Y.), 1963, 1, 341.
17. Gavosto, F., Pileri, A. and Brusca, A., *Biochem. Biophys. Acta,* 1957, 24, 250.
18. Knox, W. E. and Greengard, O., *Weber Advances in Enzyme Regulation* (Pergamon Press, N.Y.), 1965, 3, 247.

HISTOCHEMICAL LOCALIZATION OF Δ^5-3β-, 17β- AND 11β-HYDROXYSTEROID DEHYDROGENASES AND GLUCOSE-6-PHOSPHATE DEHYDROGENASE ACTIVITIES IN THE TESTIS OF BAT, *VESPERUGO PIPISTRELLUS* (DOBSON)

WITH the development of new histochemical techniques that detect the steroidogenic cellular sites in a steroid synthesizing organ, there have been a number of papers dealing with the demonstration of Δ^5-3 β-hydroxysteroid dehydrogenase (Δ^5-3 β-HSDH), 17 β-hydroxysteroid dehydrogenase (17 β-HSDH), 11 β-hydroxysteroid dehydrogenase (11 β-HSDH) in the vertebrate gonads and adrenals[1]. However, among Chiropteran mammals such studies are limited to only one species, *Myotis lucifugus*[1]. The present paper deals with the histochemical localization and significance of Δ^5-3 β-HSDH, 17 β-HSDH, 11 β-HSDH and glucose-6-phosphate dehydrogenase (G–6–PDH) enzyme activities in the testis of a Chiropteran species, *Vesperugo pipistrellus* obtained from surrounding areas of Dharwar. The histochemical procedures were similar to those described earlier[1,2].

All the enzyme activities occurred in the interstitial Leydig cells (Fig. 1) with varying intensity (Table I). A trace enzyme activity also occurred in the seminiferous epithelium.

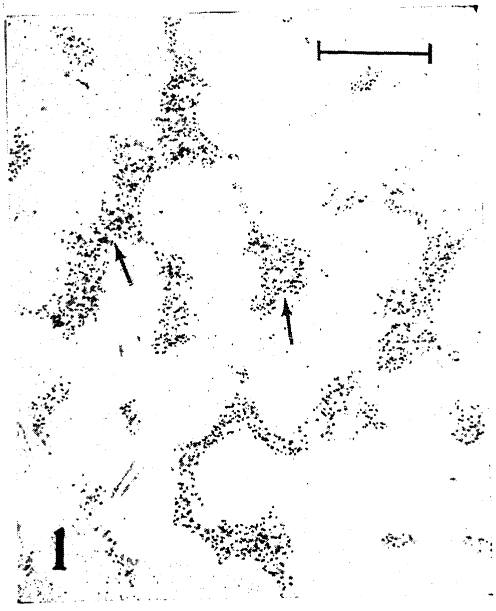

FIG. 1. Transverse section of testis of *V. pipistrellus* showing intense Δ^5-3 β-HSDH activity in the interstitial Leydig cells (arrows) and a trace enzyme activity in the seminiferous epithelium. Scale line indicates 100 mμ.

TABLE I

Enzyme Activity and the Substrate used	Intensity of reaction[1] Leydig cells	Intensity of reaction[1] Seminiferous epithelium
1. Δ^5–3 β–HSDH (Pregnenolone, Dehydroepiandrosterone)	+++	+
2. 17β–HSDH (Estradiol–17β, Testosterone)	+	+
3. 11β–HSDH (11β–Hydroxyandrostenedione)	+	+
4. G–6–PDH (disodium salt of D–glucose–6–phosphate)	+++++	++
5. Control	–	–

[1] Intensity of reaction is graded from (+) to (+++++); (+) denotes slight reaction and (+++++) a maximal reaction.

The enzyme Δ^5-3 β-HSDH is known to play a key role in the early biosynthesis of all biologically active steroid hormones[1]. The occurrence of this enzyme activity in the Leydig cells and to some extent in the seminiferous epithelial cells of *V. pipistrellus* suggests their capacity to synthesize steroid hormones. Earlier, this enzyme activity was reported in the bat *M. lucifugus* testis[1]. The 17 β-HSDH enzyme is principally involved in the biosynthesis of sex steroids and is known to catalyze testosterone-androstenedione and estradiol-17 β-estrone interconversions. The occurrence of this enzyme in the same cells as that of Δ^5-3 β-HSDH suggests that the bat testis is capable of synthesizing both androgens as well as estrogens (the sex steroids). 11 β-Hydroxysteroid dehydrogenase is principally involved in corticosteroidogenesis and is capable of converting 11 β-hydroxysteroids to 11-ketosteroids. The presence of this enzyme in the Leydig and seminiferous epithelial cells suggests the possible metabolism/synthesis of 11 β-hydroxyandrogens, 11 β-hydroxyestrogens and 11 β-hydroxycorticosteroids, in the bat *V. pipistrellus* testis. Further, as regards the histochemical demonstrations of 17 β-HSDH and 11 β-HSDH in the testis of Chiropterans is concerned, the present report seems to be the first, to the best of the author's knowledge.

The occurrence of intense G–6–PDH activity in the same eclls as that of Δ^5-3 β-HSDH, 17 β-HSDH and 11 β-HSDH in the bat testis provides additional, albeit indirect, evidence of steroidogenic potentiality of these cells, for, G–6–PDH is known to generate NADPH (pentose phosphate pathway) needed for hydroxylations during steroidogenesis[3].

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January 27, 1976.

---

1. Baillie, A. H., Ferguson, M. M. and Hart, D. McK., In: *Developments in Steroid Histochemistry*, Academic Press, London, 1966.
2. Saidapur, S. K. and Nadkarni, V. B., *Indian J. Exp. Biol.*, 1972, 10, 425.
3. Wiest, W. G. and Kidwell, W. R., In: *The Gonads* (K. W. McKerns, ed.), Appleton-Century-Crofts, New York, 1969, p. 295.

DISTRIBUTION OF GLYCOGEN AND PHOSPHORYLASE ACTIVITY IN THE VARIOUS MUSCLES OF FROG

THE study of different types of muscles has shown that the red and white fibers differ not only in colour but also in the physiological function, histology, histochemistry and energy metabolism[1]. White muscles are capable of rapid but brief contractions and mostly utilize glycolysis for energy production, whereas red muscles, which contract for prolonged periods, rely chiefly on oxidative metabolism[1]. Intermediate fibers have a moderately high oxidative capacity and low glycogenolytic activity and twitch slowly. Cardiac muscle mostly depends upon aerobic metabolism for its energy requirements[2].

Earlier investigations[3] have shown that phosphorylase activity is higher in white muscle than in the red. Present work aims at elucidating the glycogen levels and phosphorylase activity of various types of muscles in frog.

Material and Methods

The phasic sartorius, tonic ileofibularis, mixed gastrocnemius[4], and cardiac muscles of *Rana hexadactyla* were isolated with least injury from a freshly pithed animal.

Glycogen was isolated and estimated by the Anthrone method[5]. Activities of phosphorylases *a* and *ab* were estimated in the direction of glycogen synthesis[6] as described earlier[7].

Results and Discussion

Glycogen content showed variation in the four types of mucles in accordance with their physiological behaviour. Glycogen is rapidly mobilized, and is a labile energy fuel and hence its content reflects the metabolic nature in relation to physiological function of the muscle[3,7,10]. The gastrocnemius muscle showed the highest value for glycogen. The slow muscle had higher glycogen content as compared to the fast muscle (Table I). This is understandable since the fast muscle is involved in brief, rapid contractions, and the metabolism is geared towards glycogenolysis for meeting the energy requirements[3]. Cardiac muscle showed lesser glycogen content than slow and mixed muscles. Even though glycogen content is reported to be lowest in the heart muscles of air-breathing vertebrates[9] the fast muscle, in the present study, interestingly showed lesser glycogen content than the cardiac muscle. Variations in glycogen level and metabolism are dependent on the balanced activities of enzymes involved in the breakdown and synthesis of glycogen[10].

TABLE I

The levels of glycogen (mg/gm wet weight), phosphorylase a *and* ab *(μ moles of Pi/mg protein/hour) in fast, slow, mixed and cardiac muscles of* Rana hexadactyla

(Values are mean ± SD of at least 6 observations)

Sl. No.	Tissue	Glycogen	Phosphorylase *a*	Phosphorylase *ab*
1.	Sartorius (fast)	$2\cdot48 \pm 1\cdot0^*$	$62\cdot8 \pm 9\cdot1^*$	$137\cdot48 \pm 4\cdot5^*$
2.	Ileofibularis (slow)	$5\cdot67 \pm 1\cdot8^*$	$43\cdot2 \pm 9\cdot7^*$	$105\cdot6 \pm 10\cdot5^*$
3.	Gastrocnemius (mixed)	$8\cdot82 \pm 1\cdot1^*$	$52\cdot7 \pm 7\cdot8$	$101\cdot0 \pm 10\cdot1$
4.	Cardiac	$3\cdot25 \pm 0\cdot19^*$	$30\cdot11 \pm 6\cdot9^*$	$57\cdot6 \pm 6\cdot3$

* Significant at $p < 0\cdot01$ level.

The trend for phosphorylase activity was however different from that observed for glycogen content. Data presented indicate that the activity is in the order fast > mixed > slow > heart muscles (Table I). This difference in phosphorylase activity, which is the rate controlling step in the glycogenolytic pathway[3], display the different energy metabolizing capacity of the muscles in accordance with their energy demands. White muscle, capable of only short, intense bouts of anaerobic work, is known to be oriented towards glycolysis and glycogenolysis and the red muscle, which shows sustained contractions for long periods, is mainly dependent on oxidative metabolism for energy purposes[1,3,11]. Hence, phosphorylase activity is higher in white muscle, as compared to the red muscle. Cardiac muscle. which mainly depends on the blood-borne substrates, showed less phosphorylase activity as compared to the other muscles.

Department of Zoology, P. PADMAVATHI.
S.V. University, K. S. SWAMI.
Tirupati 517 502, *December* 10, 1975.

1. Szent-Gyorgyi, A., *Chemical Physiology of Contraction in Body and Heart Muscle,* Academic Press, New York, 1953.
2. Baldwin, K. M., Winder, W. W., Terjung, R. L. and Holloszy, J. O., *Am. J. Physiol.,* 1973, 225, 962.
3. Beatty, C. H. and Bocek, R. M., In : *Physiology and Biochemistry of Muscle as a Food,* Vol. II, Eds. E. J. Briskley, R. G. Cassens and B. B. Marsh, University of Wisconsin Press, 1970.
4. Kuffler, S. W. and Vaughan-Williams, E. M., *J. Physiol.* (London), 1953, 121, 589.
5. Carrol, N. V., Longley, R. M. and Roe, J. H., *J. Biol. Chem.,* 1956, 220, 583.
6. Cori, G. T., Illingworth, B. and Keller, P. G., In : *Methods in Enzymology,* Vol. I, Eds.. S. P. Colowick and N. O. Kaplan, Academic Press, New York, p. 220.
7. Padmavathy, P., Satyanarayana, K. and Swami, K. S., *Curr. Sci.,* 1975, 44, 575.
8. Larner, J., *Intermediary Metabolism and its Regulation,* Prentice-Hall, 1971
9. Bass, A., Chari, N. and Hajek, I., *Physiol. Bohemoslov.,* 1972, 21, 19.
10. Harper, H. A., *Review of Physiological Chemistry,* Lange Medical Books, California, 1973.
11. Reis, D. J., Wooten, G. F. and Hollenberg, M., *Am. J. Physiol.,* 1967, 213, 592.

HISTOCHEMICAL NATURE OF THE TRANSLATOR APPARATUS IN *PERGULARIA DAEMIA* (FORSK.) McC. AND BLAT.

THE microspores of each anther-sac in *Pergularia daemia* (Asclepiadaceae) are aggregated to form the pollinium. There are five pairs of pollinia, each pair having been derived from two adjacent anther halves. The two pollinia of each pair are joined in the middle to the corpusculum by short retinacula (*r*; Fig. 1 A). The corpusculum with two retinacula constitute the translator apparatus (or the translator). The corpusculum has a cap-like main body with two lateral blades. The translator is characteristically adapted for effective insect dispersal of the pollinia[1]. Little is known about the histochemical nature of the translator. The present communication reports the histochemical nature of the translator in *Pergularia daemia.*

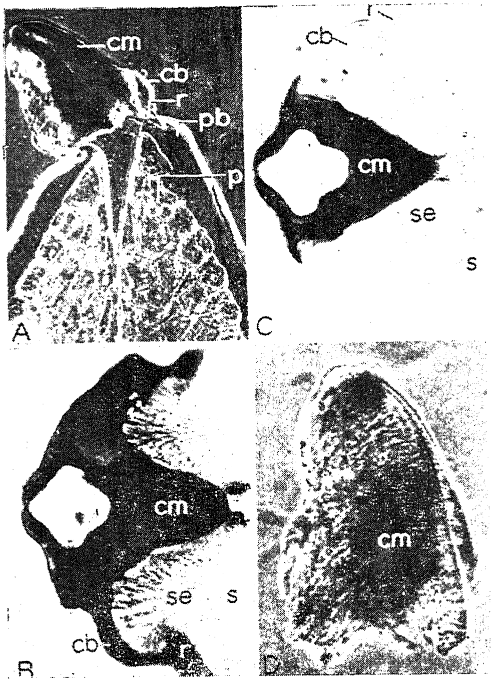

FIG. 1 A–D. *Pergularia daemia.* A and D, in phase contrast optics ; B and C, in normal optics. A. Wholemount of translator apparatus showing various parts (× 57). B. Transection of corpusculum stained with saturated solution of Sudan black B in 70% aqueous ethanol to show the presence of lipoidal compounds (× 124). C. Transection of corpusculum stained with 2% ethanolic ferric chloride, to show the presence of phenolic groups in the main body of corpusculum (× 125). D. Wholemount of corpusculum after acetolysis. (× 120). (*cb* = blades of the corpusculum ; *cm* = main body of the corpusculum ; *p* = pollinium ; *pb* = beak of the pollinium ; *r* = retinaculum ; *s* = stigma ; *se* = stigmatic epidermis.)

Investigations were performed on the material fixed in 4% formaldehyde, in 0·1 M phosphate buffer at pH 7·0, dehydrated in an ethanol series and embedded in paraffin wax. Localization of total insoluble polysaccharides was done by periodic acid/Schiff's (PAS) reaction[2]; general proteins by mercury bromophenol blue (MBB) and naphthol yellow S (NYS) tests[2]; lipids by Sudan dyes[3]; lignin by azure B, Schiff's reagent and phloroglucinol/HCl tests[3]; and phenolic radicals by ethanolic ferric chloride[4]. The translators were also observed through a Zetopan-Binolux II, dark-field fluorescence microscope. Freshly dissected translators were treated in the acetolysis mixture[5]. Extraction for lignin was done in dioxan–HCl mixture[6]. Methanolic potash procedure was followed for the selective disintegration of cutin[6]. The translators were also treated with several lipid elution techniques[3,6].

There is no evidence for the presence of insoluble polysaccharides in the translator. The corpusculum and retinacula stain with Sudan dyes (Fig. 1 B), a characteristic response of lipids[3], cutin[2] and sporopollenin[7]. The main body of the corpusculum responds positively to azure B and Schiff's tests for lignin. Sporopollenin is also known to react like lignin during several cytochemical tests[8]. Therefore the positiveness of the main body of corpusculum to azure B and Schiff's reagents suggests that it contains sporopollenin-like substance and lignin. However, sporopollenin, unlike lignin, gives a negative reaction to phloroglucinol/HCl test. The translator does not respond to phloroglucinol/HCl test and is totally resistant to HCl/dioxan elution procedure for lignin. Only the main body of the corpusculum resists acetolysis (Fig. 1 D). Substances resistant to acetolysis are known to be sporopollenin, lipid, lignin, cutin or suberin[9]. With the exception of lipids, these metabolites are autofluorescent[9-12]. The main body of the corpusculum, like lipids, is acetolysis resistant, sudanophilic but non-autofluorescent (present work). The resistance of the corpusculum to several lipid elution procedures suggests that its lipids are in a stabilized state. In addition, the main body also possesses phenolic radicals accessible to ethanolic ferric chloride (*see* Fig. 1 C). The lateral blades of the corpusculum and retinacula disintegrate during acetolysis treatment but are sudanophilic and autofluoresce dull-white—suggestive of cutin. Like the latter[13], their material is strongly birefringent. However, the lateral blades do not respond to the methanolic-potash procedure, which usually extracts cutin. This indicates that cutin is also in a resistant and stabilized state. Only the retinacula of the translator respond positively to protein tests. Thus the cutinous retinacula appear to be impregnated with protein which may explain the well-known[1] hygroscopic nature of the retinacula.

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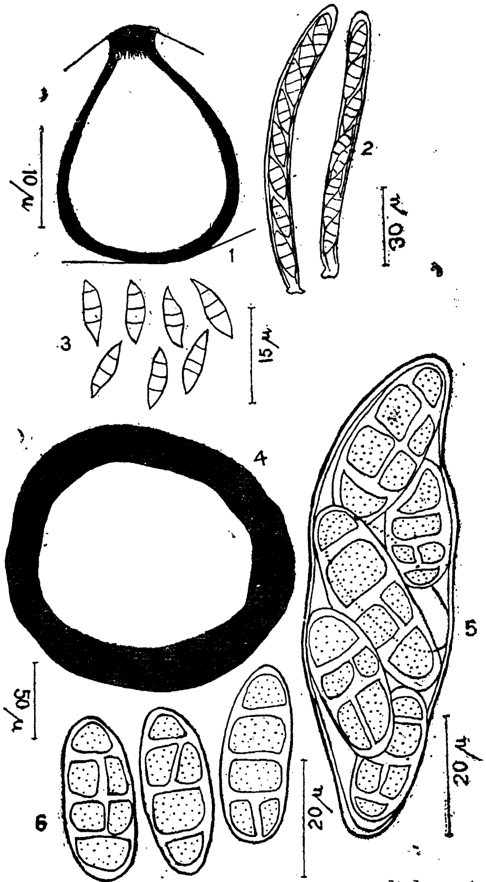
1. Macior, L. W., *Bull. Torrey bot. Club,* 1965, 92, 114.
2. Ruthmann, A., *Methods in Cell Research,* G. Bell and Sons, London, 1970.
3. Jensen, W. A., *Botanical Histochemistry,* Freeman, San Fransisco, 1962.
4. Schneider, F. L., *Qualitative Organic Microanalysis,* Academic Press, Inc., New York, 1964.
5. Erdtman, G., *Handbook of Palynology : An Introduction to the Study of Pollen Grains and Spores,* Hafner Publishing Co., New York, 1969.
6. Southworth, D., *Am. J. Bot.,* 1974, **61**, 36.
7. Ford, J. H., *In* : Brooks, J., Grant, P. R., Muir, M., van Gijzel, P. and Shaw, G. (eds.), *Sporopollenin,* Academic Press, London, 1971, p. 130.
8. Southworth, D., *J. Histochem. Cytochem.,* 1973, 21, 73.
9. Cave, C. F. and Bell, P. R., *Ann. Bot.,* 1974, 38, 17.
10. Van Gijzel, P., *In* Brooks, J., Grant, P. R., Muir, M., van Gijzel, P. and Shaw, G. (eds.), *Sporopollenin,* Academic Press, London, 1971, p. 659.
11. Chayen, J., Bitensky, L. and Butcher, R. G., *Practical Histochemistry,* John Wiley and Sons, London, 1973.
12. Goldschmidt, O., *In* Sarkanen, K. V. and Ludwig, C. H. (eds.), *Lignin : Occurrence, Formation, Structure and Reactions,* Wiley-Interscience, New York, 1971, p. 241.
13. Martin, J. T. and Juniper, B. E., *The Cuticle of Plants,* Edward Arnold, Ltd., London, 1970.

LEPTOSPHAERIA DOLIOLUM (PERS. *EX* FR.) CES. AND de NOT AND *LEPTOSPHAERULINA TRIFOLII* (ROST.) PETR.—TWO NEW HOST RECORDS FROM INDIA

DURING our studies on Ascomycetes we recorded two fungi *Leptosphaeria doliolum* (Pers. ex Fr.) Ces. and de Not, which is reported for the first time whereas *Leptosphaerulina trifolii* (Rost.) Petr. on *Helianthus annuus* L., a new host record from India. All attempts failed to culture these fungi in the laboratory. These are most probably growing saprophytically on the above hosts. *Leptosphaeria doliolum* (Pers. ex Fr.) Ces. and de Not (Figs. 1–3).

Pseudothecia scattered, superficial, hemispherical, 442–494 × 403–455 μ black with a distinct round base. Pseudothecial wall black, hard, shining made up of dense parenchymatous cells 58–87 μ in diam. Asci numerous, cylindrical, short stalked, bitunicate 96–117 × 7·5–10·5 μ, eight spored, separated by pseudoparaphysis 120–150 μ in length. Ascospores 15–21 × 4·5–6 μ in diam., brown, uniseriate, fusiform, 3 septate, end cells pointed, straight or curved, slightly constricted near the septum and 2nd cell from the apex enlarged.



FIGS. 1–6. Figs. 1–3. *Leptosphaeria doliolum*, 1. A single pseudothecium. 2. Asci with ascospores. 3. Ascospores. Figs. 4–6. *Leptosphaerulina trifolii*. 4. A single pseudothecium. 5. Ascus with ascospores. 6. Ascospores.

The above fungus differs from the original description of Lucas and Webster (1967) on *Urtica dioca* L. in having no concentric grooves at the upper part of the pseudothecium and in the size of asci.

Collected on dead wood from Pehalgaon, Kashmir (J. and K.) in October, 1972. Deposited at C.M.I., Kew, England, as I.M.I. No. 175628 and at I.A.R.I., New Delhi as H.C.I.O. No. 31903.

Leptosphaerulina trifolii (Rost.) Petr. (Figs. 4–6).

Perithecial spots 0·5–1·5 mm in diam., dark brown, black at maturity which later enlarge and coalesce forming irregular patches, 80–156 × 68–140 μ, surrounded by a thick layer of pseudothecium made up of parenchymatous cells. Pseudothecial wall 4·5–12·5 μ in diam. Asci clavate to cylindrical, thick walled, bitunicate, eight spored, 64–70 × 24–28 μ. Paraphysis and paraphysoids lacking. Ascospores 18–32 × 7–12 μ in diam.

Collected from the leaves of *Helianthus annuus* L. from Poona, Maharashtra in October, 1972. Deposited at C.M.I., Kew, England, as I.M.I. No. 190112 and at H.C.I.O. No. 31902.

The above description of the fungus tallies with the original description as given by Satya and Rajalakshmy (1964).

Sincere thanks are due to Dr. Anthony Johnston, Director, C.M.I., Kew, England, for the kind help in the confirmation of the above fungi and Dr. S. P. Raychaudhuri, for his encouragement.

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---

1. Lucas, M. T. and Webster, "Conidial states of British species of *Leptosphaeria*," *Trans. Br. mycol. Soc.*, 1967, 50, 85.
2. Satya, N. N. and Rajalakshmy, V. K., "*Leptosphaerulina trifolii* (Rostr.), Petr., a new record for India," *Curr. Sci.*, 1964, 33, 409.

A NEW STRAIN OF ALFALFA MOSAIC VIRUS FROM *PRIMULA OBCONICA*

DURING the survey for Primula strain of alfalfa mosaic virus (AMV)[1], some plants of Primula (*Primula obconica*, Hance), showing slight stunting of growth and a few scattered mild chlorotic diffused spots around leaf veins, were observed. The causal virus was found sap transmissible and produced typical symptoms of AMV[1], *i.e.*, dark brown lesions on *Phaseolus vulgaris*, L. var. Pinto (Fig. 1). But it did not infect the other two susceptible plants, *i.e.*, *Nicotiana glutinosa*. L. and *Datura stramonium*, L.

The virus culture was maintained on Primula plants. Its standard inoculum was prepared in 0·1 M phosphate buffer by the usual method. Healthy seedlings of different host plants were inoculated with virus using carborundum (400 mesh) as an abrasive. The inoculated plants were washed with water and kept in the glass-house (13–30° C) for about a month for development of symptoms. Symptomless plants were checked by back inoculation on bean plants. Electron-microscopy of the virus was done with partially purified virus preparation using carbon coated grids stained with 1% phosphotungstic acid.

The virus produced dark brown to brick red diffused necrotic lesions of varying size and shape, and often extending through veins on the inoculated leaves of *Phaseolus vulgaris*; varieties Prince, Bountiful, Kentucky Wonder, Pinto, Red Kidney and Scotia; *Vigna sinensis* (Torner) Savi and *Vigna sesquipedalis,* (L.) Wight. Necrotic lesions of varying colour and sizes were observed on *Chenopodium amaranticolor,* Coste and Reyn, *Chenopodium quinoa,* Willd, *Gomphrena globosa,* L., *Atriplex hortensis,* L., *Spinacea oleracea,* L., *Nicandra physaloides,* (L.) Gaerten, *Nicotiana debneyi,* Domin, *Nicotiana rustica,* L. and *N. tabacum,* L. (White Burley). Mild chlorotic rings developed on *Zinnia elegans,* Jacq. Young Primula plants developed small faint chlorotic spots around leaf veins only which also disappeared in the later stage of the plant growth.

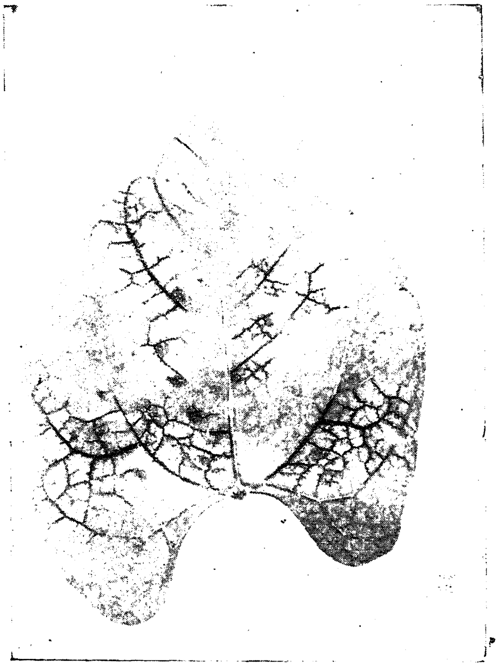

FIG. 1. AMV symptom on *P. vulgaris* var. Pinto.

The virus did not infect *Datura stramonium,* L., *Datura stramonium* var. *tatula* (L.) Torr, *Datura metel,* L., *Nicotiana glutinosa,* L., *Lycopersicon esculentum,* Mill., *Solanum tuberosum,* L., *Solanum melongena,* L., *Capsicum annuum,* L., *Capsicum pendulum,* Willd., *Petunia hybrida,* Vilm., *Cucumis sativus,* L., *Pisum sativum,* L., *Epilobium* sp.; *Brassica oleracea,* L., *Gladiolus* sp., *Beta vulgaris,* L., *Impatiens balsamina,* L., *Tagetes erecta,* L. and several wild species of *Solanum.*

The virus could withstand 1 : 600 dilution, 55–60° C temperature for ten minutes and longevity *in vitro* for 3 and 16 days at room (12–25° C) and low (5–6° C) temperatures, respectively. It also remained infective in dried leaves of *P. vulgaris* kept in desiccator for 2 and 5 months at room and low temperatures, respectively.

The electronphotomicrograph revealed the spherical shaped virus particles with an average diameter of 30 nm (Fig. 2).

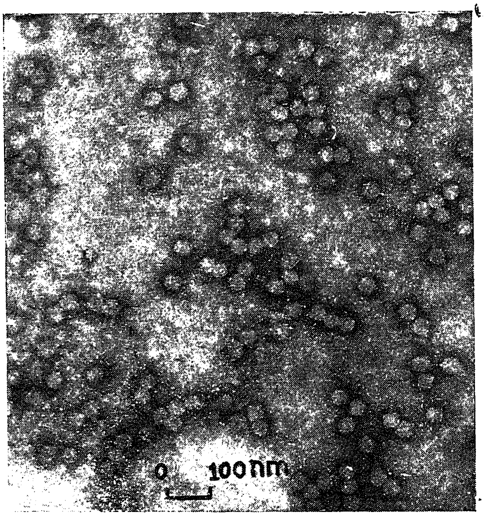

FIG. 2. Electron micrograph showing spherical particles of AMV, × 75,000.

The virus under the present study on the basis of its symptoms on bean varieties and other hosts, physical properties and particle morphology, has been identified as a strain of AMV resembling closely with its Primula strain[1]. However, it considerably differs from the Primula strain in its symptomatology on Primula and failure to infect *S. tuberosum, N. glutinosa* and *D. stramonium* plants which are normally susceptible to the latter strain. On the other hand, *N. physaloides* and *Zinnia elegans* which are resistant to Primula strain were proved susceptible to this virus. The present virus is, therefore, considered as a strain of AMV hitherto unreported.

Thanks are due to Shri M. L. Puri of Defence Science Laboratory, Delhi, for doing the electronmicroscopy of the virus.

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1. Nagaich, B. B. and Giri, B. K., *Indian Phytopath.,* 1968, 21 (4), 446.

FURTHER STUDIES ON THE EFFECT OF CHEMICALS, INTERFERING WITH PROTEIN AND NUCLEIC ACID SYNTHESIS, ON THE PROCESS OF VERNALIZATION

Chakravarti and Devi[1] suggested a positive correlation between protein and nucleic acid synthesis on the one hand and acquirement of vernalization status on the other. In the present investigation, effects of 8-azaguanine, ethionine, 5-fluorouracil and 2-thiouracil, extensively used in photoperiodic studies[2], have been determined on the vernalization of *Eruca sativa* L. culture 6518 and *Linum usitatissimum* L.R. 17.

Technique followed has been the same as in the previous study[1]. Seeds of *Eruca* and *Linum*, vernalized for 2 and 3 weeks respectively, were sown on 20–10–1975. In the present investigation, a record of both the age of the plants at anthesis and also of the number of leaves on the main axis, below the first flower, has been indicated.

Table I

Effect of certain chemicals on the vernalization of Eruca sativa *L. and* Linum usitatissimum *L.*

Chemical	Conc. ppm.	*Eruca*				*Linum*			
		Anthesis (days)	Increase over water V	Leaf No.	Increase over water V	Anthesis (days)	Increase over water V	Leaf No.	Increase over water V
Azaguanine	100	47·7	14·7*	8·5	1·1	54·1	23·3*	31·6	− 0·8
	10	39·2	6·2**	7·8	0·4	44·2	13·4*	34·0	1·6
Ethionine	500	47·8	14·8*	10·1	2·7	55·7	24·9*	48·2	15·8*
	100	45·7	12·7*	8·4	1·0	44·2	13·4*	37·2	4·8
	10	44·5	11·5*	8·8	1·4	40·0	9·2**	29·5	− 2·9
Fluorouracil	500	..	..	..	..	56·3	25·5*	45·6	13·2*
	100	47·4	14·4*	13·7	6·3*	46·0	15·2*	38·6	6·2**
	10	38·4	5·4**	12·8	5·4*	40·1	9·3**	38·5	6·1**
Thiouracil	100	49·2	16·2*	13·6	6·2*	55·4	24·6*	50·6	18·2*
	10	42·3	9·3**	11·8	4·4**	48·7	17·9*	48·6	16·2*
Water vernalized (V)		33·0	..	7·4	..	30·8	..	32·4	..
Unvernalized		50·2	17·2*	14·8	7·4*	58·7	27·9*	62·5	30·1*

* Significant at 1% level. ** Significant at 5% level.

Data presented in Table I reveal that both in *Eruca* and *Linum*, all the 4 chemicals have diminished the effect of chilling as determined by the time taken for anthesis but that is not so when the criterion of leaf number is taken into consideration. This has remained completely unaffected in azaguanine treated plants. In ethionine treatment, only *Linum* at 500 ppm has shown significant increase in the number of leaves over water vernalized. In fluorouracil and thiouracil treatments, however, this has happened in both the plants. As it is necessary that both the criteria of anthesis and node (leaf) number should be altered in the same direction for the process of vernalization to be specifically involved[3], out of the 4 chemicals, fluorouracil and thiouracil fully satisfy the requirements. They are known to inhibit photoperiodic floral induction in a large number of plants through inhibition of RNA synthesis[2]. As they also nullify low temperature effect in *Eruca* and *Linum*, there is a possibility of nucleic acid synthesis for their acquirement of vernalization status. These observations are in conformity with those of Chakravarti and Devi[1].

As regards disagreement in the results obtained through anthesis and node number in azaguanine and ethionine treatments, it is not possible to offer an explanation at this stage. However, there are other similar instances on record[4]. Before coming to a final conclusion, it would be necessary to study the effect of these chemicals on a number of other vernalizable plants.

This piece of investigation has been carried out under the U.G.C. scheme for utilization of services of retired teachers, in which one of us (S. C. C.) is participating.

Department of Botany,
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Bhopal, *January* 20, 1976.

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V. R. G. Kurup.

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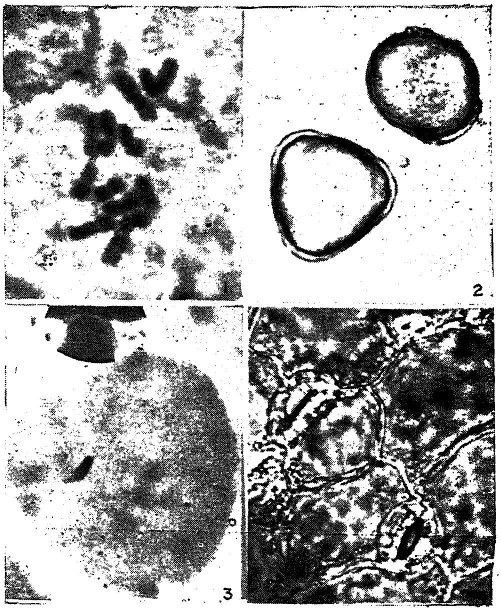
1. Chakravarti, S. C. and Devi, B. Poorna Sri, *Curr. Sci.*, 1974, 44, 58.
2. Evans, L. T. (Ed.). *The Induction of Flowering*, Macmillan of Australia, 1969.
3. Napp-Zinn, K., (Ed.), In : *Temperature and Life*, Springer-Verlag, Berlin, 1973, p. 171.
4. Chakravarti, S. C., *Proc. Nat. Acad. Sci. India*, 1964, 34 B, 216.

CHROMOSOME NUMBER AND MORPHOLOGY OF *SESBANIA PROCUMBENS* (ROXB.) W. AND A.

Sesbania Scop.. (FABACEAE) is represented by four species in India[1], namely, *S. aegyptica* Pers., *S. aculeata* Pers., *S. procumbens* (Roxb.) W. and A., and *S. grandiflora* Pers. Gamble[5] has erected *S. aculeata* var. *paludosa* of Baker as a distinct species, *viz.*, *S. paludosa*. The chromosome numbers $2n = 12$ in *S. aegyptica*[6,8], *S. aculeata*[8] and $2n = 24$ in *S. aculeata*[6] and *S. grandiflora*[7,10] have been reported earlier. The chromosome number in the only herbaceous species, *S. procumbens*, has not been reported and the same is reported here as $2n = 12$ (Fig. 1). There is no work on this rare sesban which has been recorded for the first time from a non-costal region. Morphological, cytological, palynological and anatomical characters are also presented to supplement the distinctive features of this taxon. Baker[1] and Gamble[5] have given a detailed morphological description of the species.

This species is confined to coromandel regions of peninsular India occurring chiefly in rice fields[1,3,5,9]. The material investigated by the present authors was discovered for the first time in Karnataka State, near Kukkanahalli Tank (Mysore), on 20-9-1975 (Bhaskar and Lakshmi Devi, 574 MGM) on the marshy tank bund. This also forms an addition to the 'Hydrophytes and marsh plants of Mysore' reported earlier[2].

Sesbania procumbens (basionym = *Aeschynomene procumbens* Roxb.) and *Aeschynomene indica* are usually confused since they are similar in habit and bear small sensitive leaflets and more or less yellow flowers, However, the present study has revealed many differences. *Sesbania* has a long septate fruit and diadelphous androecium (9 + 1) instead of a lomentum and diadelphous stamens in bundles of 5 each characteristic of *Aeschynomene* and a few other related genera. *S. procumbens* can be easily distinguished in the field by its bright yellow flowers while they are pale yellow or cream-coloured with red streaks in *Aeschynomene*. The stipules are absent in the latter and are small triangular and caducous in *S. procumbens*.



Figs. 1–4.

Plants in *Sesbania procumbens* flowered from August to October and fruits persist till November. The flowers were found to open in the afternoon, between 1·30 to 2·00 p.m. Pollen grains are 3-colpate (rarely 4), triangular in polar view (Fig. 2) and exine is finely reticulare. LO pattern reverse or OL (pitted ?). As in *S. aculeata* the vegetative nucleus was completely obscure at the shedding stage of pollen (Fig. 3). They were found to be mononucleate throughout the process of maturation of pollen from the smallest flower bud. However, at only one stage of the bud, *i.e.* 3 days before opening, a minute dot-like faintly stained body was observed in the pollen which is believed to be the vegetative nucleus. This becomes completely obscure very soon. A similar condition was also seen in *S. aculeata.* Pollen germinated on the stigma of the same flower, and it is monosiphonous.

The seeds which are smooth and shining germinated within five days after implanting. Stem is fistular and pitheaceous at base. Leaves are amphistomatic and stomata are anomocytic (Fig. 4) as in *S. aculeata.*

$2n = 12$ and 24 are the only reported chromosome numbers in the genus[4] and the present count in *S. procumbens* also conforms to the basic number $x = 6$ reported in the genus by earlier authors.

The authors are grateful to Prof. B. A. Razi, for his unfailing encouragement and facilities. They are also also grateful to Dr. M. A. Rau, Retd. Scientist, for his valuable suggestions. The senior author (VB) was supported by the C.S.I.R., Senior Research Fellouship.

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1. Baker, J. G., In : *Hooker Fil. Flora Brit. India,* 1876, 2, 115.
2. Bhaskar, V. and Razi, B. A., *Hydrophytes and Marsh Plants of Mysore and Surrounding Regions,* Prasaranga, Mysore Univ., 1973.
3. Cooke, T., *The Flora of Presidency of Bombay,* 1958, (Rep.), 1, 350.
4. Darlington, C. D. and Wylie, A. P., *Chromosome Atlas of Flowering Plants,* London, 1955.
5. Gamble, J. S., *The Flora of Presidency of Madras,* 1915, 1, 323.
6. Hague, (*cf.* Darlington and Wylie, 1955).
7. Jacob, (*cf.* Darlington and Wylie, 1955).
8. Rao, (*cf.* Darlington and Wylie, 1955).
9. Roxburgh, W., *Flora Indica,* 1832, 3, 337.
10. Tjio, (*cf.* Darlington and Wylie, 1955).

SOME OBSERVATIONS ON PEPPER SPOT DISEASE OF GROUNDNUT

IN the rainy season crops of groundnut during 1974 and 1975, some plants were observed to be infected with pepper leaf spot caused by *Leptosphaerulina crassiasca* (Sechet) Jackson and Bell. This disease was widespread in the region and the infected leaflets were collected from Andhra and Karnataka States. *L. crassiasca* causes both scorch and pepper spot symptoms on groundnut foliage[1]. In India only scorch symptoms have been reported[2], and this appears to be the first report of pepper spot symptoms.

The spots were round to irregular, in the size range of 0·5–1·0 mm on both sides of the leaflets. The infected leaflets remain attached to the axis for about 30 days without perceptible change in the size of the spots. Sporulation does not occur on the spots when the leaflets were still attached to the branches, but ascocarps develop rapidly on the fallen infected leaflets in the field, and within a week of incubation in moist petridishes in the laboratory. The fungus showed rapid growth of mycelium and ascocarp formation within three days on PDA. However, ascospores did not develop in cultures in continuous fluorescent light, in complete darkness or in alternating light and dark conditions.

The fungus shows active dispersal mechanism, in that the ascospores eject forcibly upwards. To know the height of its ejection, infected leaflets showing well-developed ascocarps were placed in glass containers of different depths (1·5 cm, 4·0 cm, 6·0 cm, 7·5 cm and 10·0 cm) and clean dry microslides were placed so as to cover their mouths. The spores were observed on the slides kept at different heights upto 7·5 cm. The ascospores have a mucilaginous coating and they stick to the clean glass slides. They occurred as only single units and no spore clumps were observed on the slides at any height.

The ascospores, ejected on to the slides, were collected in drops of distilled water and transferred to freshly detached host leaflets in humid chambers for studying germination. The leaflets were cleared, stained and mounted after 24 hrs. Out of 500 spores, 464 had germinated (92·8%). Germtubes developed from one or more of the six celled ascospores and, in some cases, all the six cells produced them. The pattern of germtube growth is given in Table I.

The average number of germtubes from a single spore was 2·77. The number of germtubes from the two end cells is almost equal to the number of germtubes from the four middle cells. But, the average length of germtubes from middle cells is slightly higher than that of germtubes from end cells. Germtubes grow even after the formation of appresoria and thus both terminal and lateral appresoria were observed. The highest number of appresoria observed from the germtubes of a single spore was 13, and from a single germtube and its branches was five. Appresoria were formed outside the stomal region, indicating direct penetration of epidermal cells and, out of 296 appresoria observed only two were found to be formed on the stomata.

TABLE I

Pattern of germtube growth in 100 *ascospores of* Leptosphaerulina crassiasca

Germtube position	No. of spores with germtubes	Total No. of germtubes	Average length of germtubes	No. of germtubes showing branching	No. of germtubes showing appresoria	Total No. of appresoria
From the two end cells	95 (11*)	132	116 μ	60	95	129
From the four middle cells	89 (5**)	145	123·4 μ	76	107	167

* No. of spores showing germtubes from end cells only.

** No. of spores showing germtubes from middle cells only.

Air-sampling studies in groundnut fields, using a Hirst spore trap in 1974 and glass-rod sampling in 1975, revealed that the ascospores come into the air abundantly towards the end of the crop season, *i.e.,* when the leaf fall starts (September–October), although symptoms appeared in the middle of August[3]. The air borne ascospores exhibited a clear forenoon pattern of diurnal periodicity with the peak recurring at 8 a.m.

One of us (KVM) is thankful to the authorities of CSIR for awarding a Junior Research Fellowship.

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1. Jackson, C. R. and Bell, D. K., *Diseases of Peanut (Groundnut) Caused by Fungi,* University of Georgia, College of Agriculture Experiment Stations, Research Bulletin, 1969, 56, 37.
2. Nayudu, M. V., *Indian Phytopathology,* 1963, 16 (4), 384.
3. Mallaiah, K. V. and Sreeramulu, T., *Indian J. Agric. Sci.,* 1976 (In press).

ANNOUNCEMENTS

AWARD OF RESEARCH DEGREES

Utkal University, Bhubaneswar, has awarded the Ph.D. degree in Mathematics to Shri Dhruba Charan Sahoo.

Karnatak University, Dharwar, has awarded the Ph.D. degree in Botany to Shri M. S. Mahalingappa; Ph.D. degree in Chemistry to Shri Patil Basavaraj Rayanagouda.

Osmania University, Hyderabad, has awarded the Ph.D. degree in Botany to Kum. Rehana Begum; Ph.D. degree in Biochemistry to Shri V. Srinivasan.

Tamil Nadu Agricultural University, Coimbatore, has awarded the Ph.D. degree in Agriculture to Shri S. Muthuswami and Shri S. A. Radhakrishnan.

SHORT SCIENTIFIC NOTES

Pentacyclic Triterpenoids from the Stem Bark of *Zanthoxylum acanthapodium* DC

In a previous communication[1] on the lignans of the stem bark of *Zanthoxylum acanthapodium* DC we reported three unidentified compounds having melting points 174–177°, 193–195° and 119–120°. All the three compounds were thought to be lignans, but on further examination the first two compounds turned out to be pentacyclic triterpenoids.

The compound with m.p. 174–177° crystallised as white flakes from methanol, M$^+$ 424, $[\alpha]_D + 107 \cdot 6^\circ$ ($CHCl_3$). It gave positive Leibermann-Burchard and Zimmermann reactions, analysed for $C_{30}H_{48}O$ and formed a 2, 4-DNP derivative, m.p. 225–228°. The i.r., n.m.r. and mass spectral data of the compound are in agreement with those of β-amyrenone[2]. The identity was confirmed by reduction of the compound with $LiAlH_4$ in tetrahydrofuran to β-amyrin. This is the first report of the occurrence of β-amyrenone in *Zanthoxylum* species.

The compound with m.p. 193–195° was crystallised from acetone, M$^+$ 426, $[\alpha]_D + 95 \cdot 94^\circ$ ($CHCl_3$). It analysed for $C_{30}H_{50}O$ and gave a pink colour in the Leibermann-Burchard reaction. It formed an acetate, m.p 237–239°, M$^+$ 468. These properties and the i.r., n.m.r. and mass spectral data of the compound and its acetate suggested that the compound is β-amyrin[2]. Comparison with authentic samples (m.m.p. and co-t.l.c.) confirmed the identity of the compound as β-amyrin.

The third compound with m.p. 119–120° is a lignan and a study of this and other lignans forms a part of a separate communication[3].

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Waltair 530 003, *July* 24, 1976.

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1. Venkata Rao, E., Sastry, K. V. and Palanivelu, T. J., *Curr. Sci.*, 1975, **44**, 228.
2. Takashi, K., Takashi, T., Yashiko, I., Mitsnaki, K. and Itosho, *Phytochemistry*, 1971, **10**, 3308.
3. Pelter, A., Ward, R. S., Venkata Rao, E. and Sastry, K. V., *Tetrahedron* (In press).

A New Leaf Blight of Garlic Caused by *Cladosporium echinulatum* (Berk) de Vries, from Varanasi, U.P.

In 1975–76 *rabi* season garlic (*Allium sativum* L.) plants in and around B.H.U. campus were severely affected by a leaf blight caused by *Cladosporium echinulatum* (Berk) de Vries[1]. Pure culture of the fungus on inoculation on healthy leaves reproduce the disease in 10–12 days leaving no doubt about the pathogenicity of the organism. The fungus sporulates abundantly on the dead leaves and spreads to the adjacent plants. Irrigation or rainfall appear to increase the disease. In the fields visited an infection of 20–30% of the plants was quite common and in a few cases it was upto 60%.

Our thanks are due to Shri V. P. Tewari for kindly going through the manuscript and to the U.G.C. for award of Junior Fellowship to one of us (A. K. P.).

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1. de Vries, G. A., *Contribution to the Knowledge of the Genus* Cladosporium *Link* ex *Fries*, Baarn 1952, pp. 121.

Leptosphaerulina Leaf Spot of *Trifolium alexandrinum* L. A New Record in India

Trifolium alexandrinum L. locally called as '*Barseem*' is an important fodder crop of North India. During recent survey a severe disease hitherto unrecorded on this crop was observed in submountaneous areas of Hoshiarpur District (Punjab) during the month of March, 1976. During subsequent surveys it was observed widely distributed in other parts of Panjab.

The fungus of the infected material was identified as *Leptosphaerulina trifolii* (Rostr.) Petrak and the material has been deposited in C.M.I., Kew, Surrey, England, under I.M.I. No. 203299 and the Herbarium of Department of Plant Pathology, P.A.U., Ludhiana. The perithecia observed were black, long, papillate, measuring 219–390 × 130–185 μ; asci 2–5 broad and ellipsoidal, thick walled, 69–99 × 27·5–37 μ containing 8–6 ascospores in each ascus. The description of this fungus is similar to that of given by Mehrotra (1966)[1]. Earlier this fungus is reported by him from the soil but this is the first report in India on this host causing the disease on this host.

Authors acknowledge their thanks to Dr. J. S. Chohan, Professor and Head, Department of Plant Pathology, P.A.U., Ludhiana.

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July 23, 1976. H. S. REWAL.

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1. Mehrotra, B. R., *Sydowia*, 1966, **23**, 81.

On the Abnormal Carpellary Number in *Cassia fistula*

In the campus of our college, an abnormal condition of carpellary number was noticed in the flowers of *Cassia fistula*. Flowers showed bi-, tri-, tetra- and occasionally pentacarpellary condition. The incidence of abnormality was 24% of the total number of flowers. Analysis of numerical abundance of the bi-, tri-, and tetracarpellary condition (17%, 3·6% and 1·4% respectively) and statistical treatment[1] showed that the bicarpellary condition was of the highest incidence.

Failure of differentiation in the specialized parts of plants like flowers is known to be caused by pathogens such as virus[2] and mycoplasma[3-5]. The proliferation of the carpels reported here could be considered on the same basis: the "abnormal" flowers also showed "petaloid" modification of anther lobes, proliferation of petals, numerical proliferation or reduction of stamens, adnation of filaments and occasional "phyllody" of whole flowers.

The authors express their gratitude to Prof. S. Nagaiah, Principal, S.G.S. College, Tirupati and the Management of T.T. Devasthanams for encouragement.

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A. JAYARAMI REDDY.
G. LEELAKRISHNAN.
V. CHANDRASEKHARAM.

---

1. Pillai, S. K. and Sinha, H. C., *Statistical Methods for Biological Workers.* Ram Prasad and Sons, Agra-3, India, 1968.
2. Markham, R. In: *Recent Aspects of Nitrogen Metabolism in Plants.* (Eds., Hewitt, E. J. and Cutting, C. V.). Academic Press, New York and London, 1968, 203.
3. Kandaswamy, T. K., Usman, K. M., Ranganathan, K. and Ramakrishnan, C., *South. Ind. Hort.*, 1973, **21** (1), pp. 35–37.
4. —, *Hort. Abstr.*, 1973, **44**, 5953.
5. Savithri, P., Haragopal, T., Subbuswamy, B. M., Jayarami Reddy, A. and Chandresekharam, V. 1976 (In preparation).

REVIEWS AND NOTICES OF BOOKS

Dual Resonance Models. By Paul H. Frampton. (Addison-Wesley, W.A. Benjamin, Inc., Reading, Massachusetts 01867), 1974. Price: $ 22.50.

The construction by Veneziano in 1968 of the beta function expression as a model four-particle scattering amplitude was the direct inspiration behind an enormous amount of activity in elementary particle theory in the succeeding years. New ways of thinking about particles and their interactions, the dual resonance models, emerged. One of the most interesting aspects of this whole body of work is the very intricate as well as pretty mathematics that it involves. *A priori*, one would hardly have suspected that there was scope here for so much inventive and clever use of mathematical tools familiar to any quantum mechanic. The book under review gives a very complete account of these developments.

The author begins by recalling some of the ideas in particle physics phenomenology that formed the motivation for Veneziano's work. The notions of superconvergence, finite energy sum rules and duality are briefly discussed. This part is too brief for one to be able to learn about these things here for the first time, but anyway the author's intention is to recapitulate and comment on these ideas. He then introduces the Veneziano model and discusses in detail its important features. The generalisation to the N-particle case after solving the duality constraint-equations follows. A large amount of manipulation is involved in handling the various expressions and the author guides one, carefully through it all. The Koba Nielsen form for the amplitudes of the conventional dual resonance model is then developed, displaying the cyclic symmetry and projective invariance of the model amplitudes. Alternatives to the conventional model are also discussed briefly.

The oscillator operator formalism is then introduced and a thorough discussion of gauge conditions, ghost-killing mechanisms and existence of a critical space-time dimension below which alone, one has sensible physical interpretation, is given. Inocrporation of other degrees of freedom like internal symmetry and spin are treated in successive chapters. The beautiful idea of relating the operator formalism to the quantum mechanics of a relativistic string evolving in space-time is explained. The book concludes with a survey of the use of dual model amplitudes in the analysis of the data.

The theory of dual models is both interesting and rich. This book is especially complete in giving references to the literature, and anyone interested in learning about this subject could do so very well from here, provided he is prepared to supplement reading the text with a rather large dose of algebra.

N. MUKUNDA.

DNA Synthesis and its Regulations. Edited by M. Goulian, P. Hanawalt and C. F. Fox. (Published by W. A. Benjamin, Inc.), 1975. Pp. 880. Price : Not given.

This book, the third volume in a series on Molecular Biology and Cell Biology, documents the proceedings of a conference on DNA synthesis and its regulation held at Squaw Valley, California, in March 1975. A total of 56 contributions from different research workers actively engaged in DNA research are included in the book. These articles are grouped under 12 subtitles and they cover a wide range of topics. Recent developments in various aspects of DNA replication in different systems have been brought into focus in these articles. It is now clear that biosynthesis of DNA is a highly complex process which requires the participation of several enzyme systems and their organization into multicomponent complexes. As many as 10 purified enzyme preparations have been used in the synthesis of ϕ X–174 DNA in the test tube. Studies have brought to light the existence of new enzymatic activities. DNA binding protein (*p*-2), DNA unwinding protein (*p*-83) and relaxation protein (*p*-94) are of interest in this connection.

Studies in bacterial, viral and mammalian systems have well established that DNA synthesis takes place discontinuously, forming what are called Okazaki Fragments which are then joined together by ligases. The size of the fragments seems to vary in different systems. The role of RNA as initiator of DNA synthesis is now well established in different organisms. The mechanism of incorporation of ribonucleotides into DNA and its subsequent removal from DNA has been the subject of intense investigation in many laboratories. It has been shown that replication time of the chromosome in *Escherichia coli* is constant and it is independent of the doubling time of the bacterial culture.

Studies on replication of DNA in small bacterial viruses like ϕ X–174, M 13 and Mycoplasma viruses, bacterial plasmids as well as mitochondria are being carried out in various laboratories. The mechanism of replication in mitochondria seems to be somewhat different from that in the cytoplasm. Nucleic acid replication in RNA tumour viruses and study of reverse transcriptase form the topics of research in many laboratories.

The book thus brings together reports of the activities of various investigators with diverse interest in DNA replication. Except for a few brief reports most of articles are of review type. As is to be expected the style of presentation is in no way uniform. This book will be of the greatest use to those who are actively engaged in research.

J. D. C.

Solution of Problems in Soil Machanics—A problem based textbook by BHC Sutton. (Sir Isac Pitman and Sons, Ltd., Parker Street, Kingsway, London W.C. 2 B, 2 PB), 1975. Pp. xiii + 310. Price £ 3.50.

As the title suggests, this is a textbook on soil mechanics whose subject-matter is presented generally in the form of a question and answer. The book, divided into 8 chapters, covers the following aspects : physical properties and soil classification ; permeability and flow nets ; compressibility, consolidation and settlements ; shear strength ; stability of retained and free slopes, bearing capacity and stabilization of soil bases. Each chapter starts with a brief explanation of the basic concepts followed by question-answers covering derivations, explanations, test procedures and numerical work and finally ending with exercises listing a few more problems abstracted from professional papers of Institution of Civil Engineers, London, as well as Higher National Certificate and Scottish Technical Education Council examinations. It is thus evident that the book in general is consciously planned to cover a good undergraduate curriculum. However, this book does attempt to update the knowledge in this engineering science which has developed rather rapidly in recent years. The application of soil mechanics is relevant to every branch of civil engineering activity so that in a small book like the one under review, it is not possible to cover the entire practice. Yet Prof. Sutton has made commendable efforts to cover as wide a spectrum as practicable. Another noteworthy feature is the conscious effort made to give as full and a continuous exposure of the topics as possible within the framework of question-answer type of treatment. This book, besides being a good companion to students, will be a valuable addition to the design office library.

B. V. RANGANATHAM.

Complex Carbohydrates : Their Chemistry, Biosynthesis, and Functions—A set of lecture notes by Nathan Sharon. (Published by Addison-Wesley Publishing Company, Inc.), 1975. Pp. 466. Price $ 21.50.

As the author himself puts it, the field of carbohydrates was neglected by most chemists and biochemists for a long time. in fact until early 1960 s.

Since then the attitude towards this field has changed considerably in view of the findings that complex carbohydrates are not only essential for life processes but also mediate specific immunological messages as antigens and haptens, and play important role in interaction between cells through macromolecules, and as determinants of specificity on cell surfaces.

The book is based on a set of 21 lectures given by the author to graduate students. The first lecture of Introduction deals with the importance of complex carbohydrates from structural and functional points of view. Various aspects of glycoproteins, *viz.*, isolation, characterization, structural features, and biosynthesis are covered in six lectures. Separate lectures are also devoted for sialic acids, function of protein-bound carbohydrates; structure, genetic control, biosynthesis of blood group substances and mucopolysaccharides. Lastly, peptidoglycan, and lipopolysaccharides of bacterial cell wall and penicillin action are dealt in five lectures.

The information presented in this lecture notes is mainly drawn from recent review articles and original literature, and hence it is up-to-date. Pertinent references are cited at the end of each lecture notes. The text reveals lucid presentation and newer concepts. As there are no comprehensive books or monographs covering these areas, this book by Dr. Sharon is a welcome addition and would be quite useful for graduate students in biochemistry, cell biology and molecular biophysics. The reviewers have no hesitation in recommending this book for graduate students, research workers and libraries.

V. S. R. Rao.
G. Ramananda Rao.

Electronic Biology and Cancer—A New Theory of Cancer. By Albert Szent-Gyorgyi. (Marcel Dekker, Inc., 270 Madison, Avenue, New York. 10016, N.Y.), 1976. Pp. viii + 112. Price $ 13.50.

In this book Albert Szent-Gyorgyi develops the fundamentals of what he terms electronic biology supported by delightfully simple experiments. The principles are then applied to advance a new theory of cancer.

Electronic biology is the next step from molecular biology. The electronic conductivity of a protein has been found to increase by its treatment with certain electron acceptors. Szent-Gyorgyi argues that electron acceptors could unpair and mobilise the delocalised electrons of the peptide bonds of proteins, paving the way for other electronic reactions like formation of charge transfer complexes, free-radicals, electron transport chains, and photosynthesis. Further "the unbalanced forces allowed to link molecules together and integrate their actions, creating increasingly complex structures with increasingly complex and subtle functions".

Szent–Gyorgyi divides the history of life into two periods: The periods preceding and following the appearance of light and oxygen, a strong electron acceptor. In the anaerobic electron donating atmosphere of the first, the proteins must have consisted of soluble closed-shell dielectric molecules, with paired electrons and saturated energy bands. The two main functions of this period are fermentation and proliferation. This state is termed the "α-state".

This situation was changed by the appearance of oxygen, a strong electron acceptor. Desaturation of energy bands and formation of complex semisolid structures occurred. This is the β-state.

The living cell can exist reversibly either in the proliferative α-state or the static β-state. In the α-state cell division occurs. After the completion of division, the cell passes on to the β-state. Dicarbonyls like glyoxal or dehydroascorbic acid as electron acceptors could function as inhibitors of cell division.

In cancer, a cell having lost its means of passing on to the β-state finds itself perpetually in the α-state, as a consequence of various factors including mutation or viral infection. Szent-Gyorgyi cites instances of treatment with dicarbonyls resulting in the arrest of certain types of cancer.

Although the emphasis laid by the author on dicarbonyls as regulators of cell division might not meet with wide acceptance, the ideas developed in this book are novel and should interest those who are involved in cancer research.

M. Jamaluddin.

6123/76. Published by S. R. S. Sastry, Manager, Current Science Association, Bangalore-560006
and Printed at The Bangalore Press, Bangalore-560018.

SPECTRAL SHAPE OF THE $0^- \rightarrow 0^+$ BETA TRANSITION OF ^{144}Ce

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INTRODUCTION

THE shapes of beta spectra with only two axial vector matrix elements[1] contributing to the $0^- \rightarrow 0^+$ decays are of considerable interest for the theory of weak interactions. After the great success of the concept of CVC theory, the question of whether the axial-vector current is partially conserved, arose. Tadic[2] pointed out that one may expect (*a*) almost statistical shapes (*i.e.*) C (W) = constant for all $0 \rightarrow 0$ transitions with parity change and (*b*) small Fierz-type deviations from the statistical shape for allowed GT transitions, if the axial-vector current is partially conserved. It can be useful to investigate whether all the $0^- \rightarrow 0^+$ decays really have an almost statistical shape. It is the purpose of this work to analyse in detail the $0^- \rightarrow 0^+$ transitions in the decays of Ce^{144}, Pr^{144} and Ho^{166}.

There was only one measurement of the shape of the $0^- \rightarrow 0^+$ transition in ^{144}Ce by Daniel *et al.*[4]. They[4] have also subtracted the extrapolated spectrum of ^{144}Pr down to low energies by assuming the shape of the ^{144}Pr spectrum to be allowed. But the high energy component of ^{144}Pr was not well studied by that time. A systematic study[3] of the ^{144}Pr spectrum for its shape and then its use in the extrapolation down to low energies will help in determining the shape of the $0^- \rightarrow 0^+$ transition in ^{144}Ce. Hence a detailed analysis of the $0^- \rightarrow 0^+$ transition in ^{144}Pr has been undertaken. A continuous measurement down from 100 KeV to 3 MeV will be of very great accuracy rather than using the published data of C (W) for the high energy group of ^{144}Pr.

EXPERIMENTAL DETAILS AND RESULTS

Carrier free ^{144}Ce, ^{144}Pr sources were obtained from Bhabha Atomic Research Centre, Bombay, in the form of $CeCl_3$ in HCl solution in two different consignments. Uniform sources were prepared on thin aluminised mylar backing of thickness-180 $\mu g/cm^2$. The source thicknesses ranged from 25 to 35 $\mu g/cm^2$.

The Siegbahn–Slatis beta ray spectrometer used in the present investigation with a plastic well type detector is described elsewhere[5]. The mode of analysis is described in Ref. 6.

A very complete and accurate analysis of the high energy beta group in ^{144}Pr has provided the facility to study the $0^- \rightarrow 0^+$ low energy transition in ^{144}Ce.

It is possible to extend the 100% detection efficiency range of the detector used in the present work[5] down to 50 KeV by lowering the discriminator bias and raising the anode voltage. For this purpose a 6810 A RCA photomultiplier was operated at its best performance conditions with minimum discrimination bias so as to get a 100% efficiency range upto 100 KeV. At this discriminator level the photomultiplier noise was also low.

The beta spectrum was scanned from 150 KeV to 350 KeV in steps of 5 KeV and from 400 KeV to 3 MeV in steps of 25 KeV in order to get a number of points in the interested energy region to get a detailed analysis. The 3·002 MeV beta group after its correction with the observed shape correction factor is extrapolated down to 150 KeV and a subtraction analysis as described in Ref. (6) is carried out. There is an inner beta component in ^{144}Ce decay with an end-point energy of 230 KeV and with an intensity of 4·6%. The thoretical spectrum of the 230 KeV beta group was constructed with allowed shape and with an intensity of 4·6% obtained from the data sheets. This was normalised with the total beta spectrum for countrate. The shape corrected F. K. plot of the inner beta group was subtracted from the gross spectrum. The resulted spectrum was subjected to Fermi–Kurie analysis. The correct end point energy was judged from the behaviour of the shape factor plot near the end point end for a variation of a few KeV in the end point energy. The shape is analysed for shape factor coefficients with the correct end point energy 320 ± 1 KeV using the programme 'SHAPFIT'. The experimental points were found to be best fitted with a shape factor of the form C (W) = K (1 + *a* W). A weighted least-squares fit of the results has resulted in a value of *a* as *a* = −0·537 ± 0·023. This result is in good agreement with that of Daniel *et al.* The results for three runs are given in Table I. The shape factor plot for one of the runs is given in Fig. 1.

TABLE I

Shape factor results

Run No.	Eo (KeV)	'a'
1.	320 ± 1	−0·54 ± 0·02
2.	320 ± 1	−0·51 ± 0·02
3.	320 ± 1	−0·52 ± 0·02

As shown by the present investigations, the spectra of $0^- \rightarrow 0^+$ transitions of ^{144}Ce, ^{144}Pr and ^{166}Ho are not at all statistical. These results do not support the concept of a partially conserved axial vector current theory as suggested by Tadic[1].

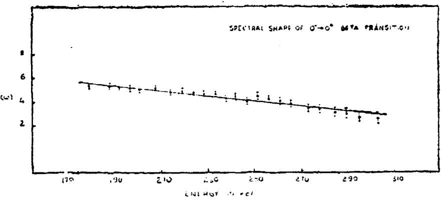

FIG. 1. Shape factor plot for the $0^- \rightarrow 0^+$ beta Transition of ^{144}Ce. Solid line is the least square fit for $C(W) = (1 + aW)$ with $A = -0.537 \pm 0.023$ and $W_0 = 320$ KeV.

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1. Nagarajan, T., Ravindranath, M. and Venkata Reddy, *Nuovo Cim.*, 1971, 3 A, 699.
2. Tadic, D., *Phys. Letts.*, 1964, 12, 116.
3. Nagarajan, T., *Ph.D. Thesis*, Andhra University.
4. Daniel, H. and Kaschl, G., *Th. Nucl. Phys.*, 1966, 76, 97.
5. Nagarajan, T., Ravindranath, M. and Venkata Reddy, K., *Nucl. Inst. Meth.*, 1969, 67, 77.
6. —, — and —, *Ibid.*, 1970, 80, 217.

SILVER (I) COMPLEXES OF SULFATHIAZOLE, SULFADIAZINE, SULFAMERAZINE AND SULFAMETHAZINE

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ABSTRACT

Silver (I)-Sulfadrug complexes of the type, AgD and Ag(HD)NO_3 where HD = sulfathiazole, sulfadiazine, sulfamerazine and sulfamethazine, have been prepared and characterized. The complexes are white, insoluble in water and organic solvents and decompose without melting well below the melting points of the drugs. The drug molecules co-ordinate in anionic or neutral form.

INTRODUCTION

In our recent publications[1-3], we have reported the preparation and characterization of the complexes of sulfadrugs with Cu (II), Zn (II), Cd (II) and Hg (II). Since the sulfadrugs are known for their bacteriostatic properties and their interactions with metal ions are not well understood, we are presently reporting their reactions with Ag (I) in aqueous and in ammoniacal media.

EXPERIMENTAL

Silver (I) chloride, nitrate and sulphate, B.D.H., A.R. Grade were used for the preparation of the complexes. Purity of the drugs was judged from their melting points and were used as such. Ag (I) chloride and sulphate complexes were prepared by adding hot ammoniacal solution of metal salt (0·01 M) to the hot solution of sulfadrug (0·01 M) in dilute ethanol keeping the drug slightly in excess while Ag (I) nitrate complexes were prepared by mixing aqueous solution of the metal-salt (0·01 M) in minimum amount of water to the hot ethanolic solution of sulfadrug (0·01 M) keeping silver nitrate in excess. The reaction mixtures were heated for about ½ hr. The complexes were filtered, washed free from the sulfadrug or silver nitrate with dilute ethanol and dried in a desiccator.

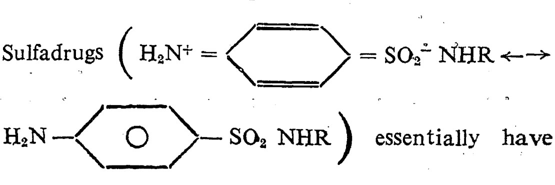
Metal part and sulphur in the complexes were estimated gravimetrically and nitrogen microanalytically, using a Coleman Nitrogen Analyzer. I.R. and electronic spectra were recorded on Perkin-Elmer-257 and Cary-14 respectively. The analytical data and some general characteristics of the complexes are summarized in Table I. Some important i.r. bands are given in Table II.

RESULTS AND DISCUSSION

The sulfadrugs form 1 : 1 complexes with Ag (1) both in water and ammoniacal solutions. The original anion is present in the complexes isolated from aqueous solution while it is absent in those isolated from ammoniacal solution. In the former case the sulfadrugs act as neutral coordinating ligand while in the latter case they act as anionic coordinating ligands. The complexes are white and insoluble in water and organic solvents. They decompose in the range 228–289° C. In the case of silver (I) chloride or sulphate complexes with drugs, the course of reaction may be as follows :

$$AgCl\ (\text{or } \tfrac{1}{2}\,SO_4^{=}) + NH_3 + HD \rightarrow Ag^+D^- + NH_4^+ + Cl^-\ (\text{or } \tfrac{1}{2}\,SO_4^{=})$$

Sulfadrugs



essentially have

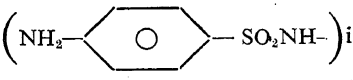
TABLE I

Analytical data and general characteristics of silver (I) complexes with sulfadrugs

Compounds	Decompn. (°C)	Found (Calc.) %			Electronic spectral bands λ_{max}.nm
		Metal	Nitrogen	Sulphur	
(*a*) Ag(ST–H)*	228 ST–200	29·29 (29·56)	11·53 (11·60)	18·05 (17·68)	275, 310 ST- 260, 300
(*b*) Ag(SD–H)*	271 SD 253	29·66 (29·97)	15·65 (15·60)	9·25 (8·96)	260, 295 SD–275, 310
(*c*) Ag(SM–H)*	276 SM–234	28·53 (28·84)	15·01 (15·09)	9·10 (8·63)	260, 300 SM–270, 305
(*d*) Ag(SZ–H)*	280 SZ–197	27·41 (27·79)	14·26 (14·54)	7·95 (8·31)	265, 310 SZ–275, 305
(*e*) Ag(ST) NO_3	237 ST–200	24·84 (25·23)	12·74 (13·20)	14·75 (15·09)	270, 305 ST–260, 300
(*f*) Ag(SD) NO_3	281 SD–253	25·78 (25·53)	16·48 (16·70)	8·00 (7·63)	270, 300 SD–275, 310
(*g*) Ag(SM) NO_3	285 SM–234	25·13 (24·71)	16·22 (16·16)	7·85 (7·39)	285, 310 SM–270, 305
(*h*) Ag(SZ) NO_3	289 SZ–197	24·12 (23·93)	15·25 (15·66)	6·70 (7·16)	260, 290 SZ–275, 305

* Prepared from Silver (I) Chloride and Sulphate.

Abbreviations : ST = Sulfathiazole, SD = Sulfadiazine, MS = Sulfamerazine, SZ = Sulfamethazine.

resonance stabilized structures[4] and the UV bands of the phenyl and SO_2 group merge to form a single strong absorption band around 260 nm[5]. In the solid state UV spectra of sulfadrugs in the present communication the maxima are shifted to longer wavelengths. The sulfadrugs in general show two bands, one due to sulfanilamide part  in the range 260–275 nm and the other due to R part in the range 300–310 nm. When the NH– proton is lost, the first band is affected while the coordination of R part affects the other band as well. If the drug acts simultaneously as an anion and coordinating ligand, both the bands are affected. This is observed in the Ag (I) complexes in the present case. The bands positions are considerably shifted from their original positions (Table I) indicating coordination of the neutral drug molecule or the drug anion to the metal.

Infrared spectra of silver (I)—sulfadrug complexes are compared with those of sulfanilamide[6] and substituted heterocyclic moieties[7] and nitrate ion[7]. Because of resonance stabilized NH_2 and NH groups in the drugs, the ν NH bands shift to lower frequencies in sulfadiazine, sulfamerazine and sulfamethazine showing the involvement of NH_2 group in the coordination. Another evidence for the coordination of NH_2 group is the negative shift of δNH_2 band between 1640–1625 cm^{-1}. The heterocyclic ring frequencies in the above drugs appearing around 1570 cm^{-1} also show negative shift of about 15 cm^{-1} indicating the coordination of ring nitrogen to the metal. In case of sulfathiazole, however, a positive shift in νNH and δNH_2 vibrations is observed but the thiazole ring frequencies at 1575 and 1532 cm^{-1} are considerably reduced in intensity due to coordination from sulphur. C–S stretching (νC–S) at 730 cm^{-1} shifts to 720 and 705 cm^{-1} in Ag (I) complexes (Nos. *a* and *e*, Table II). Out of the two SO_2-frequencies in the range 1330–1310 cm^{-1} (*νas*) and 1155–1140 cm^{-1} (ν_s), the former shows a slight negative or possitive shift while the latte remains unshifted ruling out the possibility of coordination through SO_2 group. The nitrate group frequencies at 1370 and 820 cm^{-1} correspond to unbonded nitrate[7].

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TABLE II

Some important i.r. bands of drugs and their silver (I) complexes

Drugs and Complexes	ν NH	δ NH_2	Phenyl Ring	SO_2 Asym. and Sym.	R Group	Nitrate Frequencies
ST	3320 (S) 3280 (M)	1625 (W)	1598 (S) 1500 (M)	1330 (S) 1140 (S)	1575 (S)	..
SD	3480 (S) 3380 (S)	1640 (M)	1598 (S) 1508 (M)	1310 (M) 1155 (S)	1570 (S)	..
SM	3485 (S) 3380 (S)	1640 (S)	1600 (S) 1505 (S)	1310 (M) 1155 (S)	1570 (S)	..
SZ	3440 (S) 3350 (S)	1640 (S)	1595 (S) 1510 (M)	1325 (M) 1150 (S)	1570 (M)	..
(*a*) Ag (ST–H)*	3405 (M) 3340 (M)	1640 (M)	1598 (M) 1505 (M)	1320 (M) 1140 (S)	..	..
(*b*) Ag (SD–H)*	3400 (S) 3340 (S)	1620 (W)	1600 (S) 1508 (M)	1325 (M) 1155 (S)	1560 (S)	..
(*c*) Ag (SM–H)*	3405 (M) 3340 (M)	1620 (M)	1600 (M) 1505 (M)	1330 (M) 1155 (S)	1560 (S)	..
(*d*) Ag (SZ–H)*	3425 (S) 3350 (S)	1610 (M)	1590 (S) 1510 (M)	1315 (W) 1150 (S)	1560 (M)	..
(*e*) Ag (ST) NO_3	3390 (M) 3320 (M)	1635 (M)	1595 (M) 1500 (M)	1310 (M) 1140 (S)	..	1370 (S) 820 (M)
(*f*) Ag (SD) NO_3	3370 (M) 3310 (M)	1630 (M)	1595 (S) 1508 (W)	1315 (W) 1150 (S)	1560 (S)	1370 (S) 820 (M)
(*g*) Ag (SM) NO_3	3390 (M) 3330 (M)	1625 (M)	1595 (M) 1505 (W)	1320 (W) 1150 (S)	1555 (S)	1370 (S) 820 (M)
(*h*) Ag (SZ) NO_3	3420 (S) 3350 (S)	1610 (M)	1590 (S) 1510 (M)	1310 (M) 1145 (S)	1560 (M)	1370 (S) 820 (M)

* Prepared from Silver (1) Chloride and Sulphate.

Abbreviations: ST = Sulfathiazol, SD = Sulfadiazine, SM = Sulfamerazime, SZ = Sulfamethazime.

Industrial Research, New Delhi, for the financial assistance.

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1. Narang, K. K. and Gupta, J. K., *Ind. J. Chem.*, 1975, **13**, 705.
2. —, and —, *J. Inorg. Nucl. Chem.*, 1976, **38**, 589.
3. —, and —, *Curr. Sci.*, 1976, **15**, 536.
4. Commarata, A. and Martin, A. N., *Medicinal Chemistry*, (Edited by Burger, A). Wiley—Interscience, New York, 1970, p. 119.
5. Vandenbelt, J. M. and Doub, D., *J. Am. Chem. Soc.*, 1944, **66**, 1633.
6. Nakanishi, K., *Infrared Absorption Spectroscopy*, Nankodo Company Limited (1962), p. 216.
7. Rao, C. N. R., *Application of Infrared Spectroscopy*. Academic Press, New York and London, 1963, p. 323.

SOME STABLE DICYCLOPENTADIENYL TUNGSTEN (VI) OXYDIOXIMATES AND BISINDENYL TUNGSTEN (VI) OXYDIOXIMATES

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ABSTRACT

Dicyclopentadienyl tungsten (VI) oxydichloride (DTC) and bisindenyl tungsten (VI) oxydichloride (BTC) react with some dioximes like dimethylglyoxime (DMG), α-benzildioxime (αBO) and γ-benzildioxime (γBO) in tetrahydrofuran to gave stable dicyclopentadienyl tungsten (VI) oxydioximates and bisindenyl tungsten (VI) oxydioximates respectively. The i.r. spectra, thermal stability and other properties of the products are described.

INTRODUCTION

A NUMBER of dioximes form stable chelates with metals either by replacement of the hydrogen or by the coordinating properties of the oxime nitrogen atom. DTC and BTC[1] and respective pseudohalides[2] are known but the oxydioximates of dicyclopentadienyl tungsten (VI) and bisindenyl tungsten (VI) are not mentioned in the literature. This paper describes the preparation and characterization of these compounds.

$$\underset{\text{(DTC)}}{(D)_2\,WOCl_2} + 2\,\underset{\text{(DMG)}}{C_4H_8N_2O_2} \longrightarrow$$

$$\underset{\text{(DTC}\cdot\text{2 DMG)}}{(D)_2WO\,(C_4H_7N_2O_2)} + 2\,HCl$$

$$(D)_2WOCl_2 + 2\,\underset{(\alpha\text{BO})}{C_{14}H_{12}N_2O_2} \longrightarrow$$

$$\underset{\text{(DTC}\cdot 2\,\alpha\text{ BO)}}{(D)_2WO\,(C_{14}H_{11}N_2O_2)_2} + 2\,HCl$$

$$(D)_2WOCl_2 + \underset{(\gamma\text{BO})}{C_{14}H_{12}N_2O_2} \longrightarrow$$

$$\underset{\text{(DTC}\cdot\gamma\text{ BO)}}{(D)_2WO\,(C_{14}H_{10}N_2O_2)} + 2\,HCl$$

EXPERIMENTAL

BDH grade tetrahydrofuran was purified by distillation from lithium-aluminium hydride. AR grade DMG was used in the syntheses. αBO and γBO were prepared from pure benzil as described in literature[3,4]. All reactions were carried out in dry atmosphere. I.r. spectra of the compounds were recorded on Perkin–Elmer Model-137 spectrophotometer in potassium bromide pellets. Tungsten was estimated as WO_3, carbon and hydrogen by microanalytical methods and nitrogen by semimicro-analysis.

Preparation of bis (dimethylglyoximates) and bis (α-benzildioximates)—DTC or BTC was refluxed with DMG or α BO in 1 : 2 molar ratios respectively in tetrahydrofuran. Refluxing was continued till the evolution of HCl fumes ceased. The reaction mixture was cooled and filtered through a G-4 sintered glass disc. The clear brown filtrate was evaporated to dryness under reduced pressure and the residue was washed several times with petroleum ether (60–80°) and dried. It was reprecipitated from a concentrated solution in tetrahydrofuran by petroleum ether and washed with petroleum ether and dried. Good yields were obtained in all cases. I.r. spectra of dicyclopentadienyl tungsten (VI) oxy bis (dimethylglyoximate) showed absorption in the following frequencies : 3250 s, 2950 s, 2350 m, 1700 s, 1650 sh. 1540 m, 1490 w, 1440 m, 1360 m, 1275 w, 1240 w, 1175 m, 1140 m, 1075 w, 1050 w, 960 s, 905 s, 815 m, 750 m, 705 m. I.r. spectra of bisindenyl tungsten (VI) oxy bis (α-benzildioximate) showed the following frequencies : 3450 s, 2990–2900 m (*b*), 2340 w, 1700 s, 1600 s, 1550 m, 1455 s, 1365 w, 1330 w, 1275 m, 1250 sh, 1155 m, 1080 m, 1050 sh, 950 m, 915 w, 815 w, 755 s, 715 sh, 690 m.

Preparation of γ-benzildioximates.—These compounds were prepared similarly except that the reactants were taken in molar ratios 1 : 1 and stirred for 15 hr. and refluxed for 5 hr. I.r. spectra of γ-benzildioximate of bisindenyl tungsten (VI) showed the following frequencies : 3250 sh 2960 m, 2900 m, 1710 s, 1600 m, 1540 m, 1450 m, 1375 w, 1275 w, 1175 sh, 1160 s, 1080 w, 1030 w, 1000 m, 975 m, 900 m, 825 s, 750 m, 705 w.

The compounds are pale brown solids and are quite stable under atmospheric conditions. They are insoluble in water but soluble in common organic solvents like benzene, tetrahydrofuran, acetone, etc. They do not melt upto 225° but decompose on further heating. They are attacked slowly by boiling water, hot dilute mineral acids and hot alkaline solutions. The data are summarized in Table I.

TABLE I

Data on dicyclopentadienyl tungsten (VI) oxydioximates and bisindenyl tungsten (VI) oxydioximates

Compound	Yield	Colour	Composition (%) W found (calc.)	C found (calc.)	H found (calc.)	N found (calc.)
$(C_5H_5)_2$ WO $(C_4H_7N_2O_2)_2$ (dimethylglyoximate)	76	light brown	32·75 (32·86)	38·43 (38·57)	4·32 (4·28)	9·87 (10·00)
$(C_5H_5)_2$ WO $(C_{14}H_{11}N_2O_2)_2$ (α-benzildioximate)	71	brown	22·93 (22·77)	56·27 (56·43)	4·01 (3·96)	6·88) (6·93)
$(C_5H_5)_2$ WO $(C_{14}H_{10}N_2O_2)$ (γ-benzildioximate)	60	Yellowish-brown	32·27 (32·39)	50·43 (50·70)	3·57 (3·52)	4·99 (4·93)
$(C_9H_7)_2$ WO $(C_4H_7N_2O_2)_2$ (dimethylglyoximate)	73	brown	28·03 (27·88)	47·33 (47·27)	4·17 (4·24)	8·39 (8·48)
$(C_9H_7)_2$ WO $(C_{14}H_{11}N_2O_2)_2$ (α-benzildioximate)	68	brown	20·12 (20·26)	60·51 (60·79)	4·10 (3·96)	6·09 (6·16)
$(C_9H_7)_2$ WO $(C_{14}H_{10}N_2O_2)$ (γ-benzildioximate)	69	brown	27·63 (27·54)	57·21 (57·48)	3·69 (3·59)	4·12 (4·19)

DISCUSSION

The presence of cyclopentadienyl group[5], indenyl group[6] and oximato group[7] are indicated by their usual peaks in the i.r. spectra of these compounds. The two N–O bands are probably unequal, because one hydroxyl group is ionised in the case of DMG and αBO, and different N–O linkages in the case of γ-benzildioxime in which both the hydroxyl groups are ionised. The various absorption bands in the case of bis (dimethylglyoximates) and bis (α-benzildioximates) are almost at identical positions with those of the nickel and palladium DMG complexes, indicating close structural similarities. The hydrogen bonding involved in these complexes, therefore, is not of the symmetrical type as suggested by the splitting of the O–H bands.

A distorted tetrahedral geometry has been provided for $WOCl_4$[8], $(C_5H_5)_2WOCl_2$ and $(C_9H_7)_2WOCl_2$[9] in literature. Hence it is probable that the cyclopentadienyl and the indenyl rings remain canted in all the dioximato complexes. This is indicated by the appearance of weak to medium intensity broad band in the range above 1000 cm^{-1} and medium broad band around 820 cm^{-1} [5]. In the case of dimethylglyoximato and α-benzildioximato complexes the metal atom probably acquires a seven coordination polyhedra structure (Fig. 1). The γ-benzildioximato complexes showed no absorption bands due to free hydroxyl group or hydrogen bonded hydroxyl group. The stoichiometry of the reaction and molecular formulae suggest that both the hydroxyl groups are involved in the reaction and the chelate formed is probably having a six membered ring (Fig. 2), the coordination polyhedra remains the same as in DTC and BTC.

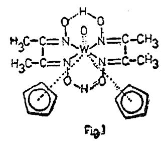

Fig1

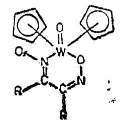

Fig2

From the data it is indicated that cyclopentadienyl and indenyl rings remain delocalized π-bonded to tungsten, while the band at ~ 960 cm^{-1} indicates the retaining of the W=O group in the prepared complexes[10].

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1. Anand, S. P., Multani, R. K. and Jain, B. D., *Curr. Sci.*, 1968, **17**, 487; *J. Organometal. Chem.*, 1969, **17**, 423.
2. Rastogi, M. K. and Multani, R. K., *J. Inorg. Nucl. Chem.*, 1975, **37**, 1995.
3. Vogel, A. I., *A Text Book of Practical Organic Chemistry*, Longman Green and Co., London, New York and Toronto, 1948.
4. Auwers, K. and Meyer, V., *Ber.*, 1899, **22**, 709.
5. Fritz, H. P., *Adv. Organomet. Chem.*, 1964, **1**, 279.
6. Scharf, G. W. and Brown, R. K., *Can. J. Chem.*, 1960, **38**, 697.
7. Blinc, R. and Hadzi, D., *J. Chem. Soc.*, 1958, p. 4536.
8. Bailar, J. C., Emeleus, H. J., Nyholm, Sir Ronald and Trotman-Dickenson, A. F., *Comprehensive Inorganic Chemistry*, 1973, p. 761.
9. Satya Pal, *Ph.D. Thesis*, Delhi University, 1972.
10. Cousins, M. and Green, M. L. H., *J. Chem. Soc.*, 1963, p. 889; 1964, p. 1967.

CUTICLE OF THE INDIAN WHIP-TAILED SCORPION *UROPROCTUS ASSAMENSIS* (STOLICZKA, 1869) AND THE SOLIFUGID *GALEODES ORIENTALIS* (STOLICZKA, 1869)

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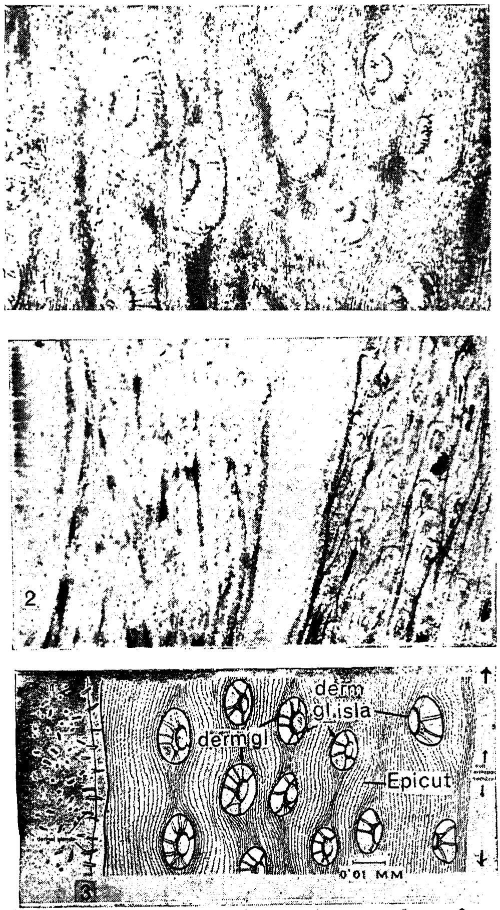
STUDIES on the structure and histochemistry of the cuticle of scorpions and spiders have been made by several workers[1-6]. The present note is an attempt to add to the knowledge of the morphology and histochemistry of some arachnids other than the scorpions and spiders. Specimens of *Uroproctus* were obtained from North-Eastern parts of India and those of *Galeodes* from the vicinity of Allahabad in Uttar Pradesh, India. Formaline-fixed, hand-cut as well as fresh frozen and paraffin sections were used for the study. Standard histochemical techniques as described by Richards[6] and Pearse[5] were followed.

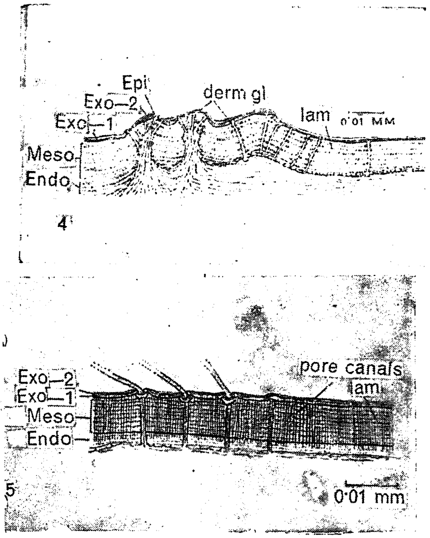
The sclerite cuticle of *Uroproctus* is chocolate-brown in colour, the external surface is granular at places and beset with tiny elevations and sparsely distributed bristles chiefly in the prosoma. The arthrodial membrane is unique in being modified into three belts each about 1·2 mm wide, beset with numerous islands of the sclerite-like cuticle of about 90 to 120 μ wide, and separated from one another by 130 μ to 170 μ of arthrodial membrane having characteristic wavy ridges on its external surface (Figs. 1 and 2). These belts are separated from one another by strips of smooth arthrodial membrane, about 0·4 mm wide without striations or ridges on its external surface. The islands are arranged in 5 to 6 rows in each strip; the gaps between the individual rows being 110–120 μ. On these islands are present a group of 4 to 5 dermal glands (Figs. 2 and 3) which appear to arise from a common pocket, *i.e.,* an outpushing of the epidermis into the mesocuticle (Fig. 4). The islands are tilted towards the side away from the tergite in all rows except the last row, *i.e.,* the row adjacent to the arthrodial membrane, where they face the opposite side. The arthrodial membrane, as usual, does not have dermal glands.

In the transverse sections of the sclerite cuticle of *Uroproctus* (Fig. 4) the outermost 2–3 μ thick exocuticle-2 of light brown colour was seen, followed by 8–9 μ thick amber coloured exocuticle-1 (nomenclature after Richards[6]), and 40–45 μ thick mesocuticle which stained red with Mallory's triple stain. In the sclerite cuticle of *Galeodes* also, the same layers were present (Fig. 5), namely, 2–3 μ exocuticle-2, 10 μ thick exocuticle-1, 80 μ thick mesocuticle and 15 μ thick endocuticle. Fine pore canals permeate all the layers of the sclerite cuticle of *Uroproctus*; these were prominent in exocuticle-1. Pore canals were very prominent in the sclerite cuticle of *Galeodes*. The sclerite cuticle of *Uroproctus* was also found to have 6–8 μ wide ducts of dermal glands opening to the outside (Fig. 4). These were simple and not united or forming groups as in the modified arthrodial membrane cuticle. Dermal glands were strikingly absent in *Galeodes*. In sections of the sclerite cuticle of *Uroproctus* stained with Mallory's, there were found 12–15 μ thick lamellae in the endocuticle and 8–9 μ thick lamellae in the mesocuticle.



FIGS. 1–3. Fig. 1. Microphotograph of the modified arthrodial membrane of *Uroproctus assamensis*, × 300. Fig. 2. Microphotograph of the arthrodial membrane of *Uroproctus* showing the dermal gland islands, × 500. Fig. 3. Diagram of the modified arthrodial membrane of *Uroproctus*.

On treating sections as well as pieces of sclerite cuticle of *Uroproctus* and *Galeodes* with concentrated nitric acid, a very thin epicuticle was resolved, which bore finger-print-like markings on the external surface. Epicuticle dissolved liberating oily droplets when strongly heated in concentrated chlorated nitric acid. Epicuticle stained blue when sections were stained with copper pthalocyanin. Positive Biuret's, Millon's, Xanthoproteic and Argentaffin reactions on the sclerite cuticle of *Uroproctus* and *Galeodes* showed aromatic plyphenols. Bleaching of the cuticle and reversal of Mallory's staining reactions in the exocuticle-1 and 2 and mesocuticle after diaphanol treatment and staining with Malachite green shows phenolic tanning of the cuticle. Negative results with thioglycolate shows absence of –S–S– bonds. Negative chitosan test of the epicuticle also showed its non-chitinous nature, while positive Argentaffin reaction and stainability with copper pthalocyanin confirmed the presence of phospholipids in the epicuticle.



FIGS. 4–5. Fig. 4. Diagram of the transverse section of the cuticle of *Uroproctus.* Fig. 5. Diagram of the transverse section of the cuticle of *Galeodes.* (Abbreviations in the diagrams : *derm. gl.,* dermal glands ; *epi. ridg.,* epidermal ridges ; *soft arthr mem.,* soft arthrodial membrane ; *epi.,* epicuticle ; *meso.,* mesocuticle ; *endo.,* endocuticle ; *exo.,* exocuticle, ; *lam.,* lamellae.)

It is evident therefore that (1) The cuticle of *Uroproctus* is unique in being modified into strips of dermal gland islands. (2) The cuticle of both of *Uroproctus* and of *Galeodes,* like those of scorpions, has a definite epicuticle. (3) The epicuticle is a non-chitinous layer and is composed chiefly of phospholipids and proteins containing aromatic groups. (4) The epicuticle of these less-known arachnids is not much different from those of scorpions and the spiders.

The author expresses his gratitude to S. C. Shrivastava, D.Sc., Reader in Zoology, University of Lucknow, under whose guidance the work was carried out.. The author is also grateful to the management of Lucknow Christian College where the work was done.

1. Krishnan, G., *Quart. Jour. Micros. Sci.,* 1953, 94, 11.
2. Krishnakumaran, A., *Zool. Jb. Anat. Bd.,* 80, 1962, S, 49.
3. Lal, M. B. and Shrivastava, S. C., *Science,* 1957, 126, 508.
4. Shrivastava, S. C., *Curr. Sci.,* 1955, **24,** 24 ; *Zool. Anz.,* 1962, **168,** 54 ; *Ibid.,* 1963, 170, 68 ; *Experientia,* 1970, 26, 1028.
5. Pearse, A. G. E., *Histochemistry,* Theoretical and Applied, J.A. Churchil, London, 1960.
6. Richards, A. G., *The Integument of Arthropods,* University of Minnesota Press, Minneapolis, 1951.

ON IDIOBLASTS IN A FEW TAXA OF PENAEACEAE

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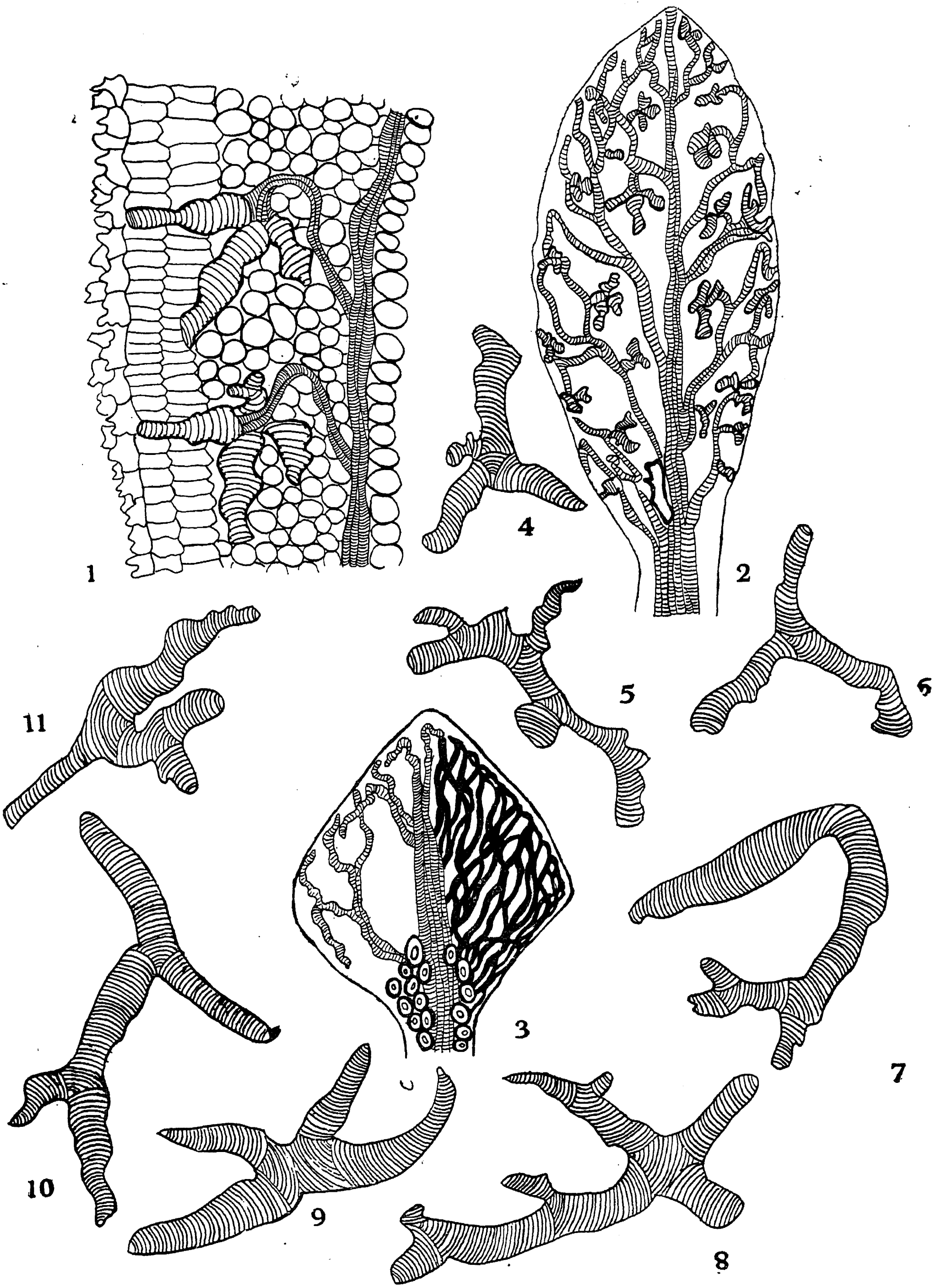
OCCURRENCE of idioblasts in the form of tracheoids, sclereids and their co-occurrence in the mesophyll at the vein-endings have opened a new interest in seed plants. The co-occurrence of filiform sclereids and wandering tracheidal veins in the mesophyll of a few taxa of this family has already been reported by a few workers[1-4]. Tracheoids, however required special attention because of their diverse size, shape, and positional relationship to the vein-endings. This aspect, studied in detail for a few taxa of this family is presented here.

MATERIALS

***Penaea**—P. mucronata* L., Africa, Burchell 394 (LE) ; Africa, Burchell 623 (LE), Eschscholts *s.n.* (LE) *P. myrtioides* L.f., Africa, Drege *s.n.* (LE) ; Royal Botanic Garden, Kew. Anon 6069 (LE) ; Africa, Drege 8155 (LE). *P. fruticulosa* L.f., Africa, Cape of Good Hope, C. Wright *s.n.* (LE) ; Echlen 3 (LE) ; Spec. *s.n.* (LE). *P.* sp. Herb. Fischer *s.n.* (LE). *Sarcocolla—S. acuta* Kth., Africa, Zeyher 3728 (LE) ; *S. furcata* Endl. Africa, Zeyher (LE) ; *S. formosa* Kth., E.E. Spec. 1838 (LE) ; *S. retzoides* Eckl and Zeyh. Africa, Burchell 7712 (LE) ; Africa, Zeyher 3724 (LE) ; *S. speciosa* Eckl. Africa, Zeyher 3725 (LE). *S.* ap., Africa, Burchell 8141 (LE).

Distributional Pattern

The singularly interesting feature in the laminae of the aforesaid taxa is the co-occurrence of sclereids and tracheoids of varied forms of exceptional interest. These idioblasts can be classified under two patterns : diffused sclereids and terminal tracheoids. Sclereids are densely scattered in the mesophyll and frequently exhibit branching at the marginal regions of the lamina. In sections cut



FIGS. 1–11. Figs. 1–3. Semi-Diagrammatic sketches. Fig. 1. Margin of lamina of *Sarcocolla retzoides* showing Lageniform terminal tracheoids. Fig. 2. Surface view of the cleared lamina of *Sarcocolla retzoides* showing loose clusters of terminal heterotracheoids of the vein-endings. Fig. 3. Surface view of the cleared lamina of *Penaea mucronata* showing filiform sclereids and terminal Loratotracheoids along the margin. Note spheroidal sclereids at base of Lamina. Figs. 4–11. Diverse types of heterotracheoids from the lamina of *Sarcocolla retzoides*, × 60 each.

parallel to midrib and passing through the marginal portion of the lamina they show intra-epidermal orientation inside the mesophyll. Their close disposition obscures the positional relationship to the vein-endings. It is presumed, however, that they are of diffuse pattern.

Tracheoids unlike the sclereids, show distinct terminal relationship with the vein-endings. As an ontogenic study is yet not carried out, the term terminal is based purely on positional relationship to the vein-endings. Both the types of idioblasts are found to occur in a few taxa but in some only tracheoids unaccompanied by any sclereids have been observed.

TYPES OF SCLEREIDS

Diffused sclereids are classified following the classification outlined by Rao and Bhupal[5].

Filiform sclereids: These thread-like cell forms are densely scattered in the mesophyll. They are quite long or curiously bent and often show branching leading to Y-shape or T-shaped cell forms. This type is seen in the lamina of *Penaea mucronata* L. (= *P. myrtiodes* L.f.), *Sarcocolla acuta* Kth., *S. formosa* Kth., *S. furcata* Endl and *S. speciosa* Eckl (Fig. 3).

Fusoid sclereids: They are observed within the vicinity of the midrib region in the leaves of *Penaea eneorum* Meerb (= *P. fruticulosa* L.f. = *Stylapterus fruticulosus* L.f.) A. Juss. and also in *Sarcocolla retzoides* Eckl and Keyh.

Spheroidal sclereids: They are present at the basal and also in the vicinity of the midrib region in the leaves of *Sarcocolla formosa* Kth. and *S. furcata Endl.* accompanied by filiform sclereids (Fig. 3).

TYPES OF TRACHEOIDS

They are distinct base forms of exceptional interest. In a few taxa they exhibit varied shape and size like the polyramous sclereids *sensu* Rao and Bhupal[5] (Figs. 1–11). A perusal of the literature shows that such a diversity of foliar tracheoids has not been recorded for seed plants. In view of this they are grouped under idiotracheoids and under this category the following two sub-types are recognised based mainly on base forms: Loratotracheoids and Heterotracheoids.

(*i*) Loratotracheoids (Fig. 3)

They are nothing but a continuation of the trachedial veinlet often found traversing the spongy or palisade region of the mesophyll especially near the marginal areas of the lamina of *Penaea mucronata, Sarcocolla acuta* and *S. furcata.* They are strap-shaped relatively thin walled, helically thickened and possess uneven width and rounded ends. The unique feature of these wandering tracheiodal veins is their independent disposition in the palisade region often found abutting the epidermal layer with more or less rounded ends.

(ii) Heterotracheoids

As a contrast to the relatively uniform tracheoids as reported in a few taxa of *Salicornia*[6] (Chenopodiaceae), *Vochysia*[7] (Vochysiaceae), *Xanthophyllum*[8] (Rutaceae), *Pogonophora*[9] (Euphorbiaceae) the tracheoids of diverse size and form closely disposed in association with veinlet endings in the laminae of *Sarcocolla retzoides* Eckl and Keyh (Burchell 7712, Zeyher 3724) and *Penaea encorum* Meerb (*P. fruticulosa* L.f. = *Stylapteris fruticulosa* A. Juss. C. Wright, *s.n.*) deserve special attention. These tracheoids are found in loose clusters at the veinlet endings and resemble Macrotracheoids *sensu* Rao and Bhattacharya[10] but differ from them in having uneven wider diameter leading to diverse shape and size in different species and in the same species (Figs. 4–11). Usually they are thin walled with spiral thickenings and protrude into the adjacent tissue prominently near the veinlet endings In transections they are found in the mid-mesophyl around the vasculature.. Along the margin of the lamina the Laganiform tracheoids were observed in *Sarcocolla retzoides* (Fig. 1).

Regarding the function of tracheoids, Van Tieghem[11] considered them as an irrigation tissue comparable to *Podocarpus* and *Cycas* and also to *Cansjera* and other members of Opiliaceae. Rao considered them to be parts of 'the tracheidal veins and Dahlgreen[4] has confirmed their connection with the veins. Thus it is clear that their close connection with the veins and veinlet endings and other structural details makes us to believe that they are probably a part of the tracheidal whole connected with conduction or storage of water.

We wish to tender our thanks to the authorities of V.I. Komarov Botanical Institute, Leningrad U.S.S.R., for the gift of leaf-specimens for our study.

1. Solereder, H., *Systematic Anatomy of th Dicotyledons,* Vols. I and II, Clarendo Press, Oxford, 1908.
2. Metcalfe, C. R. and Chalk, I., *Anatomy o Dicotyledons* Vols. I and II, Clarendo Press, Oxford, 1957.
3. Rao, T. A., *Sci. and Cult.,* 1965, 31, 380.
4. Dahlgreen, R., *Opera Botanica,* 1971, 29, 6.
5. Rao, T. A. and Bhupal, O. P., *Proc. India Acad. Sci.,* 1973, 78 B, 41.
6. De Fraine, E., *J. Linn. Soc. Bot.,* 1912, 41 B 317.
7. Rao, T. A. and Mody, K. J., *Proc. India Acad. Sci.,* 1961, 53 B, 257.
8. Dickison, W. C., *Bot. J. Linn. Soc.,* 197 67, 103.
9. Foster, A. S., *Protoplasma,* 1965, 26, 184.
10. Rao, T. A. and Bhattacharya, J., *Indian Sc Cong.,* 1976, (Abstract). Botany Section.
11. Tieghem, PH. van, *Ann. Sci. Nat. Bot.,* 189 17, 185.

LETTERS TO THE EDITOR

POLARIZATION OF FLUORESCENCE OF TETRABROMO-SODIUM FLUORESCEIN

THE fluorescence emission from tetrabromo-sodium fluorescein consists of a broad band extending from ~ 540 nm to ~ 600 nm which corresponds to a $\pi \leftrightarrow \pi^*$ transition. There are several excitation bands, corresponding to different excited states, *viz.*, the first singlet excited state S_1 $[^1(\pi\pi^*)]$, the second singlet excited state S_2 $[^1(\pi\pi^*)]$, etc., which give rise to the same fluorescence emission. In order to get information about these excited states, the polarization spectrum is investigated for different exciting wavelengths. The polarization spectrum is measured with Aminco Bowman spectrophotofluorometer at ~ 39° C for a concentration of ~ 10^{-4} g/cc of tetrabromo-sodium fluorescein in glycerol. Polarization of the emission band was measured for those excitation wavelengths for which the intensity of fluorescence emission was maximum. The effect of scattering was checked by using pure glycerol in the cell. In general, the error in the measurement of polarization is ± 1·5% but the accuracy is better in the region of the maximum emission. The results obtained are given in Table I.

TABLE I

λ_{em} nm	Percentage polarization		
	$\lambda_{ex} \sim 554$ nm	$\lambda_{ex} \sim 462$ nm	$\lambda_{ex} \sim 352$ nm
540	..	15·2	− 16·1
550	..	22·4	− 23·2
568	26·3	28·8	− 4·0
580	25·1	25·5	− 6·4
590	24·9	26·5	− 4·8
600	25·0	22·4	− 4·1

The observations indicate that there are three values of λ_{ex}, *via.*, 352 nm, 462 nm and 554 nm. The last two long wavelengths of excitation correspond to λ_{ab}. 480 nm and 510 nm reported earlier by Rathi *et al.*[1], as due to $\pi\pi^*$ and $\eta\pi^*$ transitions. The fluorescence band 540 nm to 600 nm with the maximum intensity of emission at $\lambda_{em} \sim 568$ nm corresponds to $\pi\pi^*$ transition. The $\eta\pi^*$ state possibly degrades through internal conversion to $\pi\pi^*$ state and gives rise to the same emission band. It can be seen from the observations (Table I) that the polarization is positive in both the cases of $\pi\pi^*$ and $\eta\pi^*$ excitation indicating the transition moment direction for these levels as almost parallel. However, when exciting wavelength 352 nm is used, the fluorescence is negatively polarized. It appears that with this excitation, the dye molecule is excited to the second excited singlet state S_2 $[^1(\pi\pi^*)]$ which by internal conversion gets down to the first excited singlet state S_1 $[^1(\pi\pi^*)]$ and then the molecule returns to the ground state by fluorescence emission[3-5].

These transitions agree with the suggested[2] energy levels for the molecule under consideration. The negative polarization for $\lambda_{ex} \sim 352$ nm further suggests that the transition moment directions of the first and second $^1(\pi\pi^*)$ states are somewhat normal to each other. During the time, the higher level drops to the lower with electron reorientation, the depolarization takes place and hence the magnitude of observed polarization should be less. This is in conformity with the observation, because it can be seen from Table I, that the average value of polarization for $\lambda_{ex} \sim 554$ nm is ~ 25% but for $\lambda_{ex} \sim 352$ nm it is far less ~ − 5%, for the emission band 568 nm to 600 nm.

It may also be noted that the polarization for emission wavelengths from ~ 560 nm to ~ 600 nm is almost constant, within experimental errors, for all the three excitation wavelengths. In a classical sense this corresponds to one type of emission oscillator which could mean that for this concentration (~ 10^{-4} g/cc) the dye exists in solution, only in one molecular form.

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1. Rathi, S. S. *et al., Acta Phys. Hung.*, 1968, 25, 245 ; 1969, 26, 309.
2. Machwe, M. K., *Jour. of Pure and Appl. Phys.*, 1975, 13, 337.
3. Hercules, David, M., *Fluorescence and Phosphorescence Analysis,* Interscience Publishers, 1967.
4. Feofilov, P. P., *The Physical Basis of Polarized Emission,* Consultants Bureau, New York, 1961.
5. Pesce, Rosen, Pasby, *Fluorescence Spectroscopy,* Marcel–Dekkar, Inc., New York, 1971.

A SIMPLE METHOD FOR THE EVALUATION OF FORCE CONSTANTS FROM ISOTOPIC FREQUENCIES

THE indeterminate nature of molecular force fields, when the vibration frequencies are the only available experimental data, is a well-known problem in vibrational spectroscopy. This difficulty arises due to the fact that a vibration species of order 'n' involves $\frac{1}{2} n (n + 1)$ force constant elements F_{ij} in general and their evaluation with 'n' vibration frequencies is mathematically impossible. Consequently, any attempt to evaluate the molecular force field unambiguously, should be based on the incorporation of additional experimental data on the problem.

Various methods to incorporate the isotopic frequencies as additional data, into the problem of solving the force field have been suggested by Chalmer *et al.*[1], Nibler *et al.*[2], Jordanov *et al.*[3], and Ananthakrishnan *et al.*[4] Torok[5] has shown that even in cases where a set of isotopic frequencies are available, there would be $\frac{1}{2} (n - 1) (n - 2)$ degrees of freedom still left behind, for the choice of F matrix, thus making the problem again unsolvable for all cases of $n > 2$. A method, much simpler than all the existing ones, to evaluate the force field using isotopic frequencies as additional data in the $n = 2$ cases is presented here by employing a certain parametric approach.

The general solution of the vibrational problem can be written, in the parametric framework[4,6], as

$$F = \tilde{L}_0^{-1} C \Lambda \tilde{C} L_0^{-1} \tag{1}$$

The only condition imposed on the L_0 matrix is

$$L_0 \tilde{L}_0 = G \tag{2}$$

G being the well-known inverse kinetic energy matrix[7]. Here, we employ the lower triangular form[4,6] for L_0 so that $(L_0)_{ij} = 0,\ i < j$. In the $n = 2$ case, $(L_0)_{11} = G_{11}^{1/2}$, $(L_0)_{21} = G_{12}\ G_{11}^{-1/2}$ and $(L_0)_{22} = |G|^{1/2}\ G_{11}^{-1/2}$. Λ is a diagonal matrix of elements Λ_i calculable[7] from the vibration frequencies ω_i and C is an orthogonal matrix consisting of parameters to be determined. In the $n = 2$ cases, the matrix C will consist of only a single parameter and can be written in the form[4],

$$C = \frac{1}{\sqrt{1 + c^2}} \begin{bmatrix} 1 & c \\ -c & 1 \end{bmatrix} \tag{3}$$

The force constants and hence the F matrix are invariant under isotopic substitution of atoms in the molecule. This, in turn, enables us to write eqn. (1) as

$$\tilde{L}_0^{-1} C \Lambda \tilde{C} L_0^{-1} = \tilde{L}_0^{*-1} C^* \Lambda^* \tilde{C}^* L_0^{*-1} \tag{4}$$

Here, the asterisks denote the case after isotopic substitution. Now, by virtue of the orthogonality of C^*, we can write a condition from eqn. (4) as

$$\text{Trace}\ [\tilde{L}_0^* \tilde{L}_0^{-1} C \Lambda \tilde{C} L_0^{-1} L_0^*] = \text{Trace}\ \Lambda^* \tag{5}$$

which in turn leads to a single equation of the form

$$p c^2 + q c + r = 0 \tag{6}$$

where

$$p = \frac{G_{11}^*}{G_{11}} \left(\Lambda_2 + G_{12}^2 \frac{\Lambda_1}{|G|} \right) + (G_{11} G_{22}^* - 2 G_{12} G_{12}^*) \frac{\Lambda_1}{|G|} - (\Lambda_1^* + \Lambda_2^*)$$

$$q = 2 \left(G_{12}^* - G_{12} \frac{G_{11}^*}{G_{11}} \right) \frac{(\Lambda_2 - \Lambda_1)}{|G|^{1/2}}$$

$$r = \frac{G_{11}^*}{G_{11}} \left(\Lambda_1 + G_{12}^2 \frac{\Lambda_2}{|G|} \right) + (G_{11} G_{22}^* - 2 G_{12} G_{12}^*) \frac{\Lambda_2}{|G|} - (\Lambda_1^* + \Lambda_2^*)$$

The only unknown in eqn. (6) is the parameter

TABLE I

Values of parameters and force constants

Molecule	Isotopic substitution	Values* of parameter c		Force constants (mdyn/Å)		Ref. for harmonic frequencies
		c_A	c_V	F_{ij}	Set for c_A	
Methane	H → D	0·0345	4·211	F_{11} (F_2)	5·387	9
		[0·035]	[4·25]	F_{12} (F_2)	−0·194	
				F_{22} (F_2)	0·458	
Water	H → D	−0·0275	−4·428	F_{11} (A_1)	8·356	2
		[−0·095 to 0·012]	[−4·0 to −5·25]	F_{12} (A_1)	0·192	
				F_{22} (A_1)	0·753	
Nitrogen dioxide	$^{11}N \to {}^{15}N$	0·1193	−6·045	F_{11} (A_1)	12·610	10
		[0·068 to 0·125]	[−5·0]	F_{12} (A_1)	0·431	
				F_{22} (A_1)	1·137	

* Values reported earlier are given within square brackets.

c which can be solved directly. Once c is known, F can be calculated using eqns. (3), (2) and (1).

Equation (6) being a quadratic will lead to two solutions for c (say c_A and c_V) and hence two sets of force fields fitting frequencies of the molecule and its isotopic substituent. Such a multiplicity of solutions has been reported earlier[4,8] the anomalous one corresponding to larger value of c $(= c_V)$ leading to inverse assignment of frequencies and can be eliminated by considering further data like mean amplitudes[4].

Equation (6) has been applied to a few molecules with sufficient experimental data and the values of the parameter c obtained are summarised in Table I along with their values reported earlier[4]. It may be noted that the values of c obtained from the present method lies well within the limits of c established earlier. The force constants evaluated with the present values of $c = c_A$ are also included in Table I. The superiority of the present method lies in the fact that eqn. (6) reduces the whole problem into a single equation which can be solved easily.

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Kalamassery 683 104, Cochin, *May* 24, 1976.

1. Chalmer, A. A. and McKean, D. C., *Spectrochim. Acta*, 1964, 20, 1089.
2. Nibler, J. W. and Pimental, G. C., *J. Mol. Spectrosc.*, 1968, 26, 294.
3. Jordanov, B. and Nikolova, B., *J. Mol. Struct.*, 1972, 13, 21.
4. Ananthakrishnan, T. R., Girijavallabhan, C. P. and Aruldhas, G., *Ibid.*, 1973, 16, 149.
5. Torok, F., *Acta Chim. Acad. Sci. Hung.*, 1968, 57, 141.
6. Ananthakrishnan, T. R. and Aruldhas, G., *Proceedings of All India Symposia on Physics Education and Research*, Cochin University and Kerala Physics Academy, Cochin, 1974, pp. 125–150.
7. Wilson, E. B., Decius, J. C. and Cross, P. C., *Molecular Vibrations*, McGraw-Hill, New York, 1955.
8. Ananthakrishnan, T. R. and Aruldhas, G., *Acta Chim. Acad. Sci. Hung.*, 1975, 85, 269.
9. Duncan, J. L. and Mills, I. M., *Spectrochim. Acta*, 1964, 20, 523.
10. Arakawa, E. T. and Nielson, A. H., *J. Mol. Spectrosc.*, 1958, 2, 413.

SUBSTRATE SELECTIVITY IN CATALYSIS BY ION-EXCHANGE RESINS

STUDIES involving ion-exchange resins as catalysts[1,2] are concerned mainly with the counter ions as the catalytically active species. We have studied the reaction of ketoximes in aqueous acetic acid at 30° C with the sulphonic acid ion-exchange resins, Amberlite IR-120 and Dowex 50 W–X 8, both of 8% crosslinking, as the catalysts to find out whether the resin brings about Beckmann rearrangement or oxime hydrolysis. If Beckmann rearrangement is the reaction catalysed, cyclohexanone oxime (I) as the substrate would yield caprolactam, an important commercial product for the manufacture of nylon-6. But contrary to our expectation, both the sulphonic acid resins were found to be catalytically inactive towards I and no significant reaction could be detected. When the substrate employed was acetophenone oxime (II), both the resins were found to be effective as catalysts in bringing about oxime hydrolysis to the corresponding carbonyl compound, with Beckmann rearrangement as a very minor side reaction. The quantitative aspect of the hydrolysis of II in presence of resin and the time of contact are indicated in Table I.

TABLE I

[Acetophenone oxime] = 0·07 M; Wt. of the resin = 1 gm; 30° C solvent = 30% aq. acetic acid (v/v); Time of contact = 24 hours

Resin	Yield of acetophenone*
Amberlite IR-120	19%
Dowex 50 W–X 8	24%

* Estimated by peak height comparison (GLC) with standard solution of authentic sample.

The substrate selectivity observed by us can be attributed to (*i*) preferential sorption of the aromatic substrate on the polystyrene matrix by London interactions since styrene-type ion-exchange resins sorb aromatic compounds more strongly[3-5] than aliphatic compounds of comparable molecular weight and (*ii*) the conformational freedom associated with I rendering it difficult to enter into the narrow pores of the resin where the catalytically active species, H+, are present as the counter ion.

Ion-exchange resins are crosslinked polyelectrolytes and a cation-exchange resin can be represented schematically as follows.

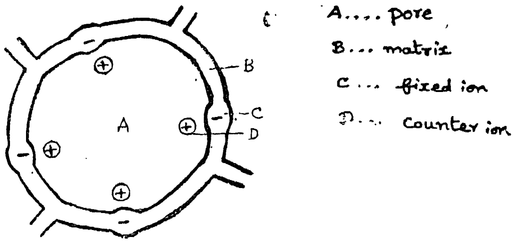

FIG. 1. Structure of a cation-exchange resin.

If the catalytic effect of an ion-exchange resin can be considered as the sum of the contributions of (1) counter-ion effect, (2) matrix effect, (3) fixed ion effect, (4) pore effect, etc., appropriate contributions from each, depending upon the nature of the substrate and the reaction would make the resin to be 'specific', despite the fact that the specificity is not so much pronounced as in the case of enzymes. In the present study, counter-ion effect would be the same towards both the substrates. Pore effect, which would be a function of the pore size and the molecular size of the substrate, cannot completely exclude I from entering the pores. But the matrix effect which may be considered to be proportional to the adsorption of the substrate on the matrix would not favour the reaction of I. In other words, the cooperative interactions of the pore and matrix effects make I to be inactive, the catalytic effect of the counter-ion not being made available to I. In the case of II, the polar end of the substrate, *i.e.,* oxime group which is the reaction centre would be held by electrostatic interactions inside the pores where H^+ ions and the dipolar solvent molecules are present, squeezing out the benzene nucleus which in turn would be held by sorption on the structurally similar polystyrene matrix by non-electrostatic forces. Both the matrix and pore effects favour the reaction of II. Resin-catalysed oxime hydrolysis of II is found to be very much faster than the homogeneous acid-catalysed hydrolysis[6]. Furthermore, the additional advantages are the substrate selectivity, case of removal of the catalyst and the possibility of sequential operations with the same catalyst.

It is apparent from the present study that a reaction in solution catalysed by ion-exchange resin exhibits the characteristics of both homogeneous as well as heterogeneous catalyses. Hammett *et al.*[7-9], from their studies on ester hydrolysis, postulated that the loss in entropy of the reactant molecule, due to some kind of fixation to the fixed ionic groups or the matrix, is an important factor, determining the catalytic efficiency of ion-exchange resins. Our experimental observation seems to be in accordance with Hammett's hypothesis. Further work, with oximes of differing molecular size, conformation and polarity, is in progress to substantiate the substrate selectivity observed by us in the present study.

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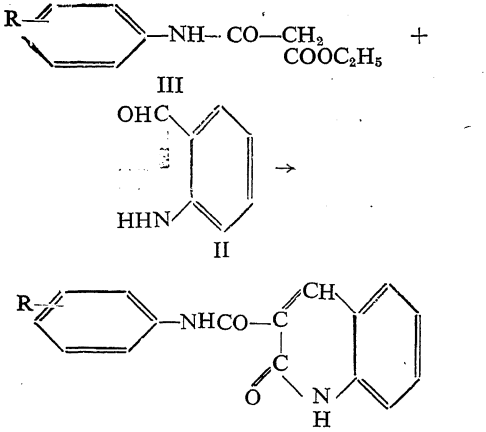
---

1. Helfferich, F., In: *Ion Exchange*, McGraw-Hill, New York, San Francisco, Toronto and London, 1962, p. 519.
2. Gauvreau, A. and Lattes, A., *C.R. Acad. Sci.,* Paris, Ser. C, 1968, 266, 1162 (Fr.).
3. Anderson, R. E. and Hansen, R. D., *Ind. Eng. Chem.,* 1955, 47, 71.
4. Chasanov, M. G., Kunin, R. and McGarvey, F., *Ibid.,* 1956, 48, 305.
5. Wheaton, R. M. and Bauman, W. C., *Ibid.,* 1953, 45, 228; *Ann. N.Y. Acad. Sci.,* 1953, 57, 159.
6. Ruzicka, L. and Wettstein, A., *Helv. Chim. Acta,* 1935, 18, 986.
7. Haskell, V. C. and Hammett, L. P., *J. Am. Chem. Soc.,* 1949, 71, 1284.
8. Bernhard, S. A. and Hammett, L. P., *Ibid.,* 1953, 75, 1798 and 5834; *Ibid.,* 1954, 76, 991.
9. Riesz, P. and Hammett, L. P., *Ibid.,* 1954, 76, 992.

o-AMINOBENZALDEHYDE CONDENSATION : FORMATION OF SOME 2-QUINOLONES

LITERATURE available on the synthesis, properties and uses of 2 and 4-quinolones is extensive[1-3]. Chakravarti[4] obtained quinolone derivatives from alkaloids. Recently Fryer *et al.*[5] prepared 2-quinolone derivatives in 50–90% yield.

This communication deals with the synthesis of some new 2-quinolones (I) by the condensation of *o*-aminobenzaldehyde (II) with some substituted malonic esters (III) like ethyl malon-*p*-iodo and *p*-bromoanilate, ethyl malon-*o*-phenetidate, ethyl malon-2 : 5-xylidate, ethyl malon-*m*- and *p*-anisidate. The ethyl esters of malon-*p*-iodoanilic acid and malon-*o*-phenetidic acid were prepared for the first time by well-known methods[6,7].



I

The I.R. spectrum of 2-quinolone shows a band at 1670 cm^{-1}, characteristic for amide band in secondary amides, mainly due to $>C=O$ in the ring and another band at 1644 cm^{-1} assigned to $>C=O$ which is not in the ring.

Experimental

Formation of ethyl malon-p-iodoanilate.—*p*-Iodoaniline (8·8 g) and ethyl malonate (9·6 g) were

TABLE I

Name	Formula	m.p. (°C)	Yield	% of N		% of C		% of H	
				Found	Calc.	Found	Calc.	Found	Calc.
1. R-*p*-Bromoanilide	$C_{16}H_{11}N_2O_2Br$	>360	88%	7·85	8·16	55·66	55·97	2·98	3·20
2. R-*o*-Phenetide	$C_{18}H_{16}N_2O_3$	244	83%	8·86	9·09	69·87	70·13	4·85	5·19
3. R-2:5-Xylidide	$C_{18}H_{16}N_2O_2$	321	90%	9·42	9·58	73·60	73·97	5·12	5·47
4. R-*m*-Anisidide	$C_{17}H_{14}N_2O_3$	296	90%	9·60	9·52	69·13	69·38	4·43	4·75
5. R-*p*-Anisidide	$C_{17}H_{14}N_2O_3$	325	91%	9·50	9·52	68·97	69·38	4·56	4·75

where R stands for 2-quinolone-3-carboxy-.

taken in a round bottomed flask and the mixture heated to very gentle ebullition with an air condensor. After one hour the mixture was cooled and 50 ml of ethanol was added. The crystals of the dianilide which separated was filtered and the filtrate concentrated in a china dish over a steam bath. The product obtained was recrystallised from petroleum ether, m.p. 115° C, yield (30%). Found: N, 3·86%; C, 39·33%; H, 3·48%; $C_{11}H_{12}NO_3I$ requires N, 4·19%; C, 39·63%; H, 3·60%.

Ethyl malon-p-*phenetidate.*—This was prepared similarly from *o*-phenetidine and ethyl malonate; yield (41·6%), m.p. 44° C. Found: N, 5·84%; C, 61·68%; H, 6·43%; $C_{13}H_{17}O_4N$ requires N, 5·58%; C, 62·15%; H, 6·77%.

Condensation of o-*aminobenzaldehyde with ethyl malon*-p-*iodoanilate: formation of 2-quinolone-3-carboxy*-p-*iodoanilide.*—Ethyl malon-*p*-iodoanilate (0·33 g), *o*-aminobenzaldehyde (0·12 g) and two drops of piperidine were heated on a steam bath for 3 hours. The mass was then washed with hot water (15 ml) and filtered. The product was insoluble in most of the common organic solvents. It was repeatedly washed with hot ethanol, m.p. > 360° C, yield (82%). Found: N, 6·92%; C, 48·98%; H, 2·46%; $C_{16}H_{11}N_2O_2I$ requires N, 7·11%; C, 49·23%; H, 2·82%.

Using the same experimental conditions, the following 2-quinolones were prepared. Melting point, percentage yield and analytical results are given in Table I.

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1. Decker, *J. Prakt. Chem.*, 1901, 64, 85.
2. Neely, A. Julius, and Capps, *J. Am. Chem. Soc.*, 1955, 77, 182.
3. Bowman, Campbell and Tanner, *J. Chem. Soc.*, 1959, p. 444.
4. Chakravarti, D. *et al.*, *Ibid.*, 1953, p. 3337.
2. Fryer, Brust and Sternbach, *Ibid.*, 1964, p. 3097.
6. Rathore, B. S. and Ittyerah, P. I., *J. Ind. Chem. Soc.*, 1960, 37, 592.
7. Singhal, O. P. and Ittyerah, P. I., *Ibid.*, 1965, 42, 480.

STABILITY CONSTANTS OF SOME TRIVALENT METAL-ION COMPLEXES OF N-BENZOYLACETONEANTHRANILIC ACID

IN this communication we are giving the dissociation constants of the ligand derived from benzoylacetone and anthranilic acid, and the formation constants of its complexes with La (III), Ce (III), Pr (III), Nd (III), Sm (III) and Gd (III) which have been determined potentiometrically (0·1 M $NaClO_4$) at 30 ± 0·1° C. The sequence of stability constants was found,

$$\text{La (III)} < \text{Ce (III)} < \text{Pr (III)} < \text{Nd (III)} < \text{Sm (III)} < \text{Gd (III)}$$

All the chemicals used were either B.D.H. AnalaR or Reidel reagents. The ligand N-Benzoylacetoneathranilic acid (abbr. H_2BA) was synthesised by the method of Pfeiffer *et al.*[1]. Calvin-Bjerrum pH-titration technique[2,3] as modified by Irving and Rossotti[4] was used for stepwise formation constant of the complexes. The ionic strength of the solution was maintained constant by $NaClO_4$ (0·1 M) solution. All the measurements were carried out at 30 ± 0·1° C.

In the case of H_2BA, the formation curve extends upto $\bar{n}_a \simeq 2·0$ indicating that two protons respectively from carboxylic and phenolic hydroxyl groups are dissociated. The values of dissociation constants as obtained by interpolation at half $\bar{n}_a$ values method and interpolation at various $\bar{n}_a$ values were found to be 9·81 and 6·20,

TABLE I

Stability constants of trivalent metal complexes of H_2BA

Temperature = 30 ± 0·1° C $\mu = 0 \cdot 1$ M $NaClO_4$

	A	B	C	D*	E*	F
La (III)						
log K_1	7·14	7·18	7·13	7·14	7·14	7·14
log K_2	3·84	3·80	3·83	3·86	3·84	3·83
log β_2	10·98	10·98	10·96	11·00	10·98	10·97
Ce (III)						
log K_2	8·56	8·59	8·58	8·56	8·56	8·57
log K_2	5·29	5·25	5·30	5·30	5·29	5·29
log β_2	13·85	13·84	13·88	13·86	13·85	13·86
Pr (III)						
log K_1	8·90	8·94	8·88	8·90	8·90	8·90
log K_2	5·65	5·61	5·68	5·70	5·65	5·66
log β_2	14·55	14·55	14·56	14·60	14·55	14·56
Nd (III)						
log K_1	9·20	9·26	9·18	9·21	9·20	9·21
log K_2	5·75	5·70	5·80	5·82	5·75	5·76
log β_2	14·95	14·96	14·98	15·03	14·95	14·97
Sm (III)						
log K_1	9·90	9·96	9·86	9·88	9·91	9·89
log K_2	6·42	6·40	6·45	6·40	6·42	6·42
log β_2	16·32	16·36	16·31	16·28	16·33	16·31
Gd (III)						
log K_1	11·18	11·20	11·16	11·20	11·18	11·18
log K_2	7·94	7·90	7·98	7·98	7·95	7·95
log β_2	19·12	19·10	19·14	19·18	19·13	19·13

A = Interpolation at various $\bar{n}$ values ; B = Interpolation at half $\bar{n}$ values ; C = Correction term ; D = Successive approximation ; E = Convergence formulae ; F = Average values.

* Most suitable methods in this case.

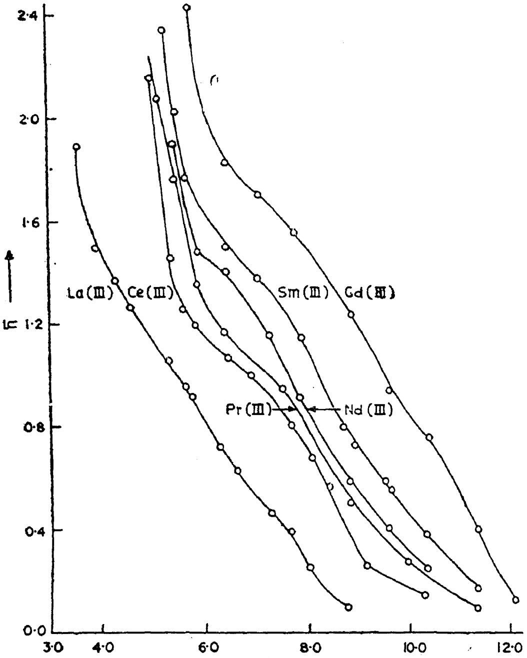

FIG. 1. Formation curves of trivalent metal—H_2BA complexes.

The formation curves (Fig. 1) of all metal-ligand systems attain maxima at $\bar{n} > 1 \cdot 5$ which indicate that both 1 : 1 and 1 : 2 complexes are formed. The values of log K_1 and log K_2 were obtained from the formation curves at $\bar{n} = 0 \cdot 5$ and $\bar{n} = 1 \cdot 5$, respectively. The values of these constants were refined by different computational methods[5] and are shown in Table I.

H_2BA being a tridentate neutralizes two equivalents of the base to yield two buffer regions in its potentiometric equilibrium,

$$H_2BA \rightleftharpoons HBA^- + H^+ ; \quad HBA^- \rightleftharpoons H^+ + BA^{--}$$

when 1 : 5 mixtures of ligand and the metal are titrated against standard alkali, the systems show inflections at lower buffer region indicating complexation. The complex equilibria may be represented by the equations,

$$H_2BA + M^{+3} \rightleftharpoons MBA^+ + 2\,H^+ ; \quad MBA^+ + H_2BA \rightleftharpoons M(BA)_2^- + 2\,H^+,$$

La (III), Ce (III), Pr (III), Nd (III), Sm (III), and Gd (III) having $4f^0$, $4f^1$, $4f^2$, $4f^3$, $4f^5$ and $4f^7$ electrons possess ionic radii, 1·061, 1·034, 1·013, 0·995, 0·964 and 0·938 Å, respectively. The stability of the complexes of the metals, having similar electronic configuration incerases with decreasing ionic size. The same trend has also been observed in the present case.

Dept. of Chemistry, P. K. KANUNGO.
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Jodhpur, India, R. K. MEHTA.
February 16, 1976.

1. Pfeiffer, P., Offermann, W. and Wermer, H., *J. Prakt. Chem.*, 1942, 159, 313.
2. Bjerrum, J., *Metal Ammine Formation in Aqueous Solution*, P. Haase and Sons, Copenhagen, 1941.
3. Calvin, M. and Wilson, K. W., *J. Am. Chem. Soc.*, 1945, 67, 2003.
4. Irving, H. and Rossotti, H. S., *J. Chem. Soc.*, 1954, p. 2904.
5. — and —, *Ibid.*, 1953, p. 3397.
6. Schroder, K. H., *Acta Chem. Scand.*, 1966, 20, 1401.

CATALYTIC ACTIVITY OF OXIDE SPINELS: DECOMPOSITION OF ISOPROPYL ALCOHOL

THE catalytic activity of spinels of the type MgM_2O_4 (M = Al, Fe, Cr) has been investigated for a variety of reactions like methanation of carbon monoxide[1], oxidation and dehydrogenation of hydrocarbons[2,3]. The catalytic properties of spinels are much dependent on their structure. In oxidation reactions, the position of the trivalent metal ion could significantly influence the reactivity of the catalyst[4]. Of the compounds $MgAl_2O_4$, $MgCr_2O_4$ and $MgFe_2O_4$, the first two are normal spinels, while $MgFe_2O_4$ is partly an inverse spinel. The purpose of the present study is to compare the activity and selectivity of the spinels in the decomposition of isopropyl alcohol in terms of the structural changes of the spinels and changes in conductivity type.

The spinels were prepared by the coprecipitation of hydroxides from the respective nitrates (BDH AR) at an optimum pH. They were then dried at 100° C and calcined at 850° C for 24 hours. The spinel formation in each case was checked by TGA, DTA and X-ray analyses. The BET (N_2) surface areas of the samples $MgAl_2O_4$, $MgCr_2O_4$ and $MgFe_2O_4$ were found to be 49·5, 19·8 and 18·3 m^2/g respectively. Isopropyl alcohol (BDH AR) was distilled before use and its purity checked by chromatograph.

The activity of the catalysts for the decomposition of isopropyl alcohol was determined in a fixed bed flow type reactor[5]. The gaseous products were analysed using Orsat's gas analyser and liquid products were estimated with a vapour phase chromatograph (Varian, 1800).

For the catalyst $MgCr_2O_4$ both dehydrogenation and dehydration reactions took place, while on $MgAl_2O_4$ only dehydration was observed to the complete exclusion of dehydrogenation with traces of condensation product, namely, diisopropyl ether ($<$ 0·5 mole %). The catalyst $MgFe_2O_4$ was predominantly dehydrogenating. The dehydration product, namely, water, in this case was very small ($<$ 1 mole %). Contact time *vs* concentration plots were made for dehydrogenation and dehydration reactions on the catalysts and from the initial slopes, the corresponding rates of reaction were calculated. These rates were used to make Arrhenius plots from which energies of activation were calculated. Table I shows the kinetic parameters for the decomposition of isopropyl alcohol on different catalysts studied.

TABLE I

Kinetic parameters for the decomposition of isopropyl alcohol

Catalyst	Rate of dehydrogenation (moles sec^{-1} m^{-2})	Rate of dehydration moles (sec^{-1} m^{-2})	Activation energy kcal/mole		Frequency factor min^{-1} m^{-2}	
			Dehydrogenation	Dehydration	Dehydrogenation	Dehydration
$MgCr_2O_4$						
285° C	0·19	0·03	11·99	16·10	$6\cdot3\times10^5$	$4\cdot8\times10^6$
310° C	0·32	0·06				
330° C	0·46	0·10				
$MgFe_2O_4$						
290° C	0·47					
310° C	0·52	..	7·14	..	$1\cdot74\times10^3$	..
335° C	0·74					
$MgAl_2O_4$						
220° C		0·14				
235° C	..	0·37	..	29·38	..	$4\cdot66\times10^{13}$
250° C		0·83				

Comparison of the dehydrogenation activities shows that the catalyst MgFe$_2$O$_4$ exhibits the highest activity followed by MgCr$_2$O$_4$, while MgAl$_2$O$_4$ does not show any dehydrogenation activity at all.

From the structural considerations, one would expect the Fe^{3+} ions situated in the tetrahedral holes of the partly inverse spinel MgFe$_2$O$_4$ to be less helpful for dehydrogenation than the Cr^{3+} ions situated in the octahedral holes of the normal spinel MgCr$_2$O$_4$. The higher activity shown by MgFe$_2$O$_4$ is probably due to its predominant *n*-type semiconductivity. The total absence of dehydrogenation activity for MgAl$_2$O$_4$ may be ascribed to the masking of the effect of Mg^{2+} ions by the highly acidic Al^{3+} ions.

Considering the dehydration step in the decomposition of isopropyl alcohol, MgAl$_2$O$_4$ exhibits the maximum activity among the three spinels and the active dehydration centres could be the surface coordinatively unsaturated Al^{3+} ions (Lewis acid sites), as in the case of Al$_2$O$_3$[6]. With MgCr$_2$O$_4$, the dehydration activity is the combined effect of a favourable *p*-type semiconductivity and to a less extent the surface coordinatively unsaturated Cr^{3+} ions (Lewis acid sites). The large difference in activation energies for dehydration between MgAl$_2$O$_4$ (29·4 kcal/mole) and MgCr$_2$O$_4$ (16·1 kcal/mole) lends support to the operation of different active centres on the catalysts. The extremely small dehydration activity observed with MgFe$_2$O$_4$ is consistent with the view that a *n*-type semiconductor does not favour dehydration reaction.

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January 31, 1976.

1. Frydenlundsevej, Vedboek, *J. Catalysis*, 1969, 14, 238.
2. Gillot, Bernard, *Bull. Soc. Chim.*, 1968, 6, 2382.
3. Anon., *C.A.*, No. 64, PC 15739 *d*.
4. Cimino, A., *La Chimica EL' Industria*, 1974, 56.
5. Pandao, S. N., Kuriacose, J. C. and Sastri, M. V. C., *J. Sci. Inastr. Res.*, 1962, 21 D, 171.
6. Krylov, O. V., *Catalysis by Nonmetals*, Academic Press, p. 131.

A NOTE ON THE LATE PLIOCENE–EARLY PLEISTOCENE OSTRACODA AND FORAMINIFERA FROM NEILL ISLAND, SOUTH ANDAMAN

THE present note deals with ostracoda and foraminifera obtained from the Sawai Bay Limestone exposed in the western coast of Neill Island. The following succession of beds (Archipelago group) has been noted in this Island :

Formations	Age	Approximate Thickness in metres
Neill Island Coral Beds	Holocene	1–3
Unconformity		
Malacca Limestone	Late Pleistocene	1–4
Sawai Bay Limestone	Late Pliocene-Early Pleistocene	3–4
Unconformity		
Sawai Bay Mudstone —Base not exposed	Late Miocene-Early Pliocene	155

The Sawai Bay Limestone has yielded a rich assemblage of ostracoda which are identified as *Bairdia antillea* van den Bold, *Bairdia* spp., *Pontocypris attenuata* Brady, *Cytheridea* aff. *Cytheridea* (*Leptocytheridea*) *sulcata* Stepenson, *Paracytheridea longicaudata* (Brady), *Caudites javana* Kingma, *Hemicytheria* sp., *Leguminocythereis* sp., *Loxoconcha tumida* Brady, *Loxoconcha* spp., *Loxoconchella honoluliensis* (Brady), *Paracytherois* sp., *Actinocythereis* sp., *Bradleya* sp., *Chrysocythere* sp., *Cythereis* sp., *Quadracythere* spp., *Xestoleberis* sp., *X. variegata* Brady, *Ruggieria* sp., *Cytherella* sp., and *Cytherelloidea atmai* Kingma. In association with this assemblage of ostracoda, the following planktonic foraminifera—*Globorotalia cultrata limbata* (Forrasini), *G. truncatulinoides* (D' orbigny), *G. tumida tumida* (Brady), *Turborotalia inflata* (D' orbigny), *Turborotalia acostaensis humerosa* (Takayanagi and Saito) and *Turborotalia acostaensis acostaensis* (Blow)—have also been recorded. This foraminiferal assemblage suggests a Late Pliocene-Early Pleistocene age to the Sawai Bay Limestone. The boundary between Pleistocene and Pliocene lies within this formation.

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1. Blow, W. H., *Proc. First Int. Conf. Plank. Micro.*. 1969, 1. 199.
2. Banner, F. T. and Blow, W. H., *Nature*, 1965, 207 (5004), 151,

EFFECT OF VARIOUS AUXINS ON GROWTH RATE OF STATIC AND SUSPENSION CULTURES OF *PHYSOCHLAINA PRAEALTA* HOOK

TISSUE culture appears to be a promising technique for future production of phytochemicals[1,2]. At this present stage of development tissue culture is not commercially viable because of the slow growth rates of plant cells and low yields of the active constituents. In this paper the authors have studied the effect of auxins on growth rate of static and suspension cultures of *Physochlaina praealta*. Growth is expressed either in terms of fresh tissue per litre per day or as growth index (G.I.), which is a value obtained by dividing the final fresh weight of tissue by its initial weight over a given period of time. Comparative accounts of large scale yields of plant cell cultures are reported in Table I[3-5].

TABLE I

Yield of cell, mass in suspension cultures

Plant	Med.	Vessel	GMS per lit per day Wet	Dry
Ginkgo, Lolium Liek	cm	Carboy	3	..
Rose	cm	Pilot plant	10	..
Carrot	cm	Carboy	6·1	..
,,	,,	Fermentor	..	1·9
Spearmint	Defined	Carboy	4·7	..
,,	,,	Flask	6·3	0·4
Rose	Defined	Phytostat	11	0·4
Hyoscyamus	..	Fermentor	1·6	0·02
Bean	Defined	Flask	..	0·7
,,	Phylone	,,	..	1·1
Kidney Bean	Defined	Fermentor	..	1·2
Apocynum	Defined	Fermentor	5·33	..
Physochlaina	Defined	Flask	19·0	2·2

It appears that *Physochlaina praealta* H. tissue grown in revised Murashige and Skoog's medium[6] with naphthalene acetic acid 0·1 ppm in flask suspension cultures has given growth rates which are faster than any other tissue reported in literature.

Static Cultures

Callus cultures were obtained from hypocotyl regions of sterile seedlings in 100 ml Erlenmeyer flasks containing 40 ml of revised Murashige and Skoog's medium supplemented with coconut water 15% and naphthalene acetic acid 1 ppm. Callus was pale white in colour and friable in nature. The callus cultures thus established and maintained by subculturing at four weeks interval were used as experimental material.

Naphthalene acetic acid, indole acetic acid and 2, 4-dichlorophenoxy acetic acid at 0·25, 0·5 and 1 ppm concentrations were used to study their effects on growth. The cultures were grown in BOD incubator in dark at 25° C. To study growth rates, ten flasks of each treatment were harvested at weekly interval over a period of eight weeks.

Effects of NAA, IAA and 2, 4-D on callus growth are presented in Fig. 1 (*a, b* and *c*). Growth index was recorded for each treatment for 6 weeks. A maximum growth yield was obtained in the media with 0·5 ppm NAA (G.I. 14·6). Growth was slightly less at 1 ppm (G.I. 11·6) while it was minimum at 0·25 ppm (G.I. 1·3). IAA from 0·25 ppm to 1 ppm produced almost uniform growth for four weeks, however further growth was maintained at 0·5 (G.I. 10·3) and 1 ppm (G.I. 9·3), while it declined at 0·25 ppm (G.I. 6·1). 2, 4-D produced maximum growth at 0·5 ppm (G.I. 12·4), while at 1 ppm (G.I. 5·5) the growth was considerably less and was minimum at 0·25 ppm (G.I. 1·8).

Suspension Cultures

Four weeks old callus tissue was transferred to 500 ml Erlenmeyer flasks containing 100 ml of liquid medium supplemented with 0·1 ppm NAA. The cultures were incubated in dark at 25° C on a rotary shaker at 120 ppm. The suspension cultures were subcultured in fresh medium at 30 days interval. A study of growth rates was conducted by harvesting four flasks at 10 days interval.

A maximum cellular fresh weight of 57 g per 100 ml medium supplemented with 0·1 ppm NAA was obtained in 30 days. The growth of fresh tissue recorded per litre per day is 19 g. which is supposed to be the highest reported value (Fig. 1 *d*).

It is observed that growth of callus in suspension cultures is faster (19 g fresh tissue/l/day, 2·2 g dry tissue/l/day), than that of static cultures (15·0 g fresh tissue/l/day, 0·68 g dry tissue/l/day).

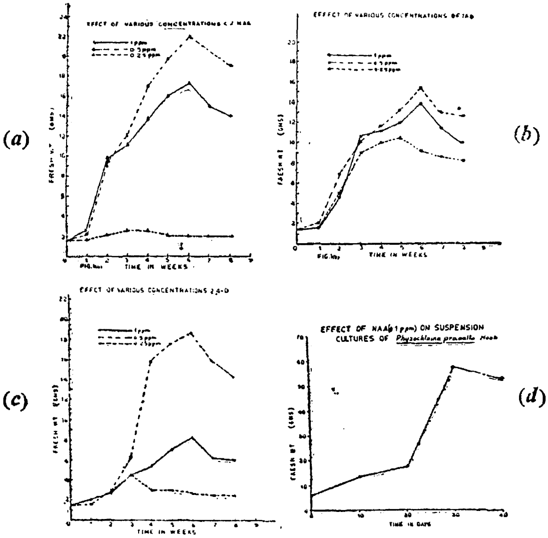

FIG. 1 (*a–d*). Fig. 1 *a–c*. Effect of various concentrations of NAA, IAA and 2,4-D on static cultures of *Physochlaina praealta*. Fig. 1 *d*. Effect of NAA (0·1 ppm) on suspension cultures of *Physochlaina praealta*.

A large quantity (2·8 kg) of fresh tissue was harvested from cultures growing on Murashige and Skoog's modified medium with NAA concentrations of 1 ppm. The tissue when analysed gave a total alkaloid value of 0·02% on dry weight basis. T.L.C. revealed that the major alkaloid formed was hyoscyamine and two minor unidentified alkaloids were also present. Hyoscine was absent.

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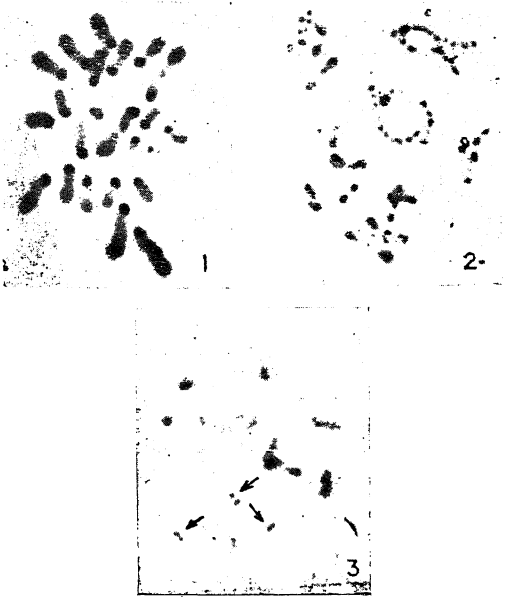
C. K. ATAL.

---

1. Staba, E. J., *Recent Advances in Phytochem.*, 1969, 75, 106.
2. Steck, W., *Planta Medica*, 1974, 25, 158.
3. Mandels, M., Mugbuire, A. and Hamed, M., *Technical Report*, 68–6–FL. Food Division, FL–58, 1967.
4. Veliky, I. A. and Martin, S. M., *Cand. J. Microbiol.*, 1970, 16, 233.
5. Lee, P. K., Carew, D. P. and Rosazza, J., *Lloydia*, 1972, 35 (2), 150.
6. Murashige, T. and Skoog. F., *Physiol. Plant.*, 1962, 15, 473.

PATTERN OF C-BANDING IN *ACRIDA TURRITA* (ACRIDIDAE—ORTHOPTERA)

THE pattern of distribution of heterochromatin in mammalian chromosomes has been extensively worked out[1,2]. But very little work has been done on the heterochromatin pattern in insect chromosomes. This communication therefore attempts to present the nature of C-banding in *Acrida turrita*.



FIGS. 1–3. Fig. 1. *Acrida turrita* spermatogonial chromosomes showing C-bands. Fig. 2. Diplotene showing distinct centromeric heterochromatin blocks. Fig. 3. Anaphase-I (one pole) showing the double nature (arrows) of the kinetochore.

Males of *Acrida turrita*, a short-horned grasshopper ($2n♂ = 23$) collected from Manasagangotri, Mysore (India) formed the material for study. Testes were dissected out and treated with hypotonic 0·9% sodium citrate solution for 45 minutes. The material was fixed in 1 : 3 acetic/methanol. Flame dried slides were prepared. C-bands were obtained by the barium-hydroxide-saline-Giemsa (BSG) technique of Sumner[3] with minor modifications. Slides were kept in saturated barium hydroxide for 15 minutes at room temperature, briefly rinsed in 0·02 N HCl and distilled water, and air dried. After incubating in 2 XSSC at 60–65° C for 3 hours in a petri dish, briefly rinsed in distilled water, stained in buffered (pH 6·8) Giemsa and mounted in DPX.

In the spermatogonial metaphase plate (Fig. 1) the C-bands can be clearly made out at the centromeric regions. The C-bands of different chromosomes are of similar size. Along with this, there are C-positive less dense telomeric blocks. The C-bands are conspicuous at the centromeric regions during diplotene (Fig. 2). The anaphase-I chromosomes also show distinct C-bands which are represented in the form of two dots (Fig. 3). The present observation on the double nature of the centromeric dots (bodies) at anaphase-I supports the earlier findings[4] that the division of the kinetochore starts at metaphase-I and accomplished at anaphase-I. This observation corroborates the heterochromatin studies on the grasshopper *Bryodema tuberculata*[5]. Further, the present studies have confirmed the telocentric nature of the chromosomes of *Acrida turrita*[6].

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---

1. Hsu, T. C. and Arrighi, F. E., *Chromosoma*, 1971, 34, 243.
2. —, *Annl. Rev. Genet.*, 1973, 7, 153.
3. Sumner, A. T., *Exptl. Cell Res.*, 1972, 75, 304.
4. Lima-de-Faria, A., *Hereditas*, 1956, 42, 85.
5. Klasterska, I., Natarajan, A. T. and Ramel, C., *Chromosoma*, 1974, 44, 393.
6. Kumaraswamy, K. R. and Rajasekarasetty, M. R. (Unpublished data).

ON THE SHEDDING OF GILL RAKER PROCESSES IN GRAY MULLETS (FAM. : MUGILIDAE)

THE possible shedding of gill raker processes in grey mullets has already been indicated[1] and in the present note, additional evidence for such shedding is indicated.

In the general teleostean plan, gill rakers are bristle-like structures in double rows, occurring along the inner margin of hypobranchial, ceratobranchial and epibranchial of the first three arches and on the fourth and fifth gill arches where the ceratobranchial of the fifth arch is further modified to form the inferior pharyngeal. Luther[1] described the morphology of the gill raker processes of the first four arches and the ceratobranchial of *M. cephalus* but the present study on *M. cephalus* showed many variations and finer details which he has not dealt with. A full description is therefore added here as further information. In the first arch, the gill rakers of the outer row are very long while those of the inner row are very short. In the second arch, the rakers of the inner row are about half in size of those of the outer row. In the third arch however they are nearly three quarters in length whilst in the fourth arch (ceratobranchial) the rakers in the inner and outer rows are equal in size. The fifth ceratobranchial (inferior pharyngeal) displays a concavity wherein parallel rows of gill rakers, equal in size, are present. The gill rakers of the first gill arch are beset with tiny processes called the primary processes in double rows on the inner face except near the distal region (Fig. 1) whereas in other arches these primary processes are present even distally (Fig. 2). On the inner sides of the gill rakers are present (excepting in the first arch)

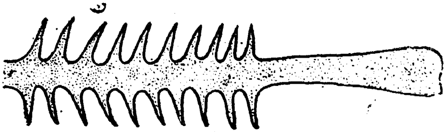
FIG.1

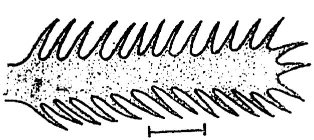
FIG.2

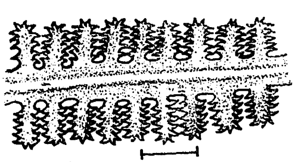
FIG.3

FIGS. 1–3. Fig. 1. Gill raker of the first gill arch with primary processes except on the distal end. Fig. 2. Gill raker of the other arches (2–5) with primary processes. Fig. 3. Strip of gill raker process with secondary and tertiary processes.

the strips, each one of which is made up of two filaments joining at the mid-dorsal line and forming a groove midventrally which fits on the gill raker. Each strip in turn bears tiny lateral processes called secondary processes and these further bear tertiary processes on both sides, thus leading to the formation of a very complex sieving apparatus (Fig. 3). Fleshy lobes were reported to be present[1] on the distal ends of the gill rakers of the third, fourth (ceratobranchial) and fifth (inferior pharyngeal) but this could not be observed in the present study.

During investigations on the feeding of mullets in Vellar estuary, gill raker processes were found to occur commonly in their stomachs. Observations on the branchial baskets of large number of mullets showed that some had loosely arranged gill raker processes while in some they were absent. Luther[1] suggested the possibility of shedding of the gill raker processes particularly from the third arch onwards, due to their clogging with copepods and detritus during the rainy period (from October to December) in Mandapam area. In order to throw more light on this, acclimatised specimens of *Mugil cephalus* and *Liza macrolepis* (10–12 cm), collected from Vellar estuary, were introduced into two aquaria containing filtered estuarine water (using bolting silk cloth No. 20) mixed with inert animal charcoal powder. After 24 hours, the water from the aquarium was filtered and the residue was washed with filtered water. When the residue was examined under the microscope, small strips of gill raker processes could be seen. The branchial basket of the experimental fish, on examination, showed an accumulation of the charcoal powder over the gill raker processes, clogging them. Strips of gill raker processes were found to be dislodged and some portions were missing. It is therefore inferred that grey mullets, under the conditions of clogging of their gill raker processes, may shed them. Clogging occurs in all the other arches excepting in the first, since the first arch is not efficient in sieving (as described earlier). According to Luther only the third, fourth and fifth arches shed the gill raker processes but presently all the arches excepting the first are found to shed them.

It should be very interesting to study the regeneration of the gill raker processes, the feeding efficiency and the physiological state of the fish after the shedding the gill raker processes.

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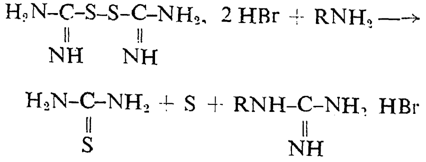
---

1. Luther, G., *J. Mar. Biol. Ass. India*, 1964, 6 (2), 251.

A CONVENIENT METHOD FOR THE PREPARATION OF MONOSUBSTITUTED GUANIDINES

VARIOUS methods for the synthesis of monosubstituted guanidines are reported in the literature but many times yields are poor and purity is not upto the mark. Also, in view of the facts, that guanidine derivatives possess high degree of biological activity[1-2], it was thought worthwhile to prepare these monosubstituted guanidines by some quick method resulting in high yields.

Bisformamidine disulphide salt on treatment with slight excess of molar proportion of primary amines afforded corresponding monosubstituted guanidines in high yields. The reaction can be depicted as follows :



$$\mathrm{H_2N{-}\underset{\underset{NH}{\|}}{C}{-}S{-}S{-}\underset{\underset{NH}{\|}}{C}{-}NH_2,\ 2\,HBr + RNH_2 \longrightarrow}$$

$$\mathrm{H_2N{-}\underset{\underset{S}{\|}}{C}{-}NH_2 + S + RNH{-}\underset{\underset{NH}{\|}}{C}{-}NH_2\ HBr}$$

where R = aryl or alkyl group.

Experimental

Bisformamidine disulphide dihydrobromide used in the synthesis of these guanidines was prepared according to the method reported in the literature[3].

Preparation of phenylguanidine.—Bisformamidine-disulphide dihydrobromide (10 g ; 0·032 mol) and aniline (4 g ; 0·043 mol) were mixed thoroughly and the reaction mixture was heated on a water bath for 3–4 hrs. The syrupy mass thus obtained was dissolved in water and then filtered. To a clear filtrate, sufficient quantity of sodium bicarbonate was added. The guanidine carbonate thus obtained was washed with ether and finally with small amounts of alcohol to remove the impurities and unreacted aniline also.

Similarly other guanidines were prepared. In some cases picrates were isolated (Table I),

TABLE I

10 g of Bisformamidine-disulphide dihydrobromide was used in each case

Sl. No.	Amines used	Name of the product	Yield %	M. pt. ° C (Derivatives)
1.	Aniline (3·2 g)	Phenyl guanidine[4]	80	138 (Carbonate)
2.	*o*-Toludine (3·8 g)	*o*-Tolyl guanidine[5]	70	134 (Hydrochloride) 223 (Picrate)
3.	*m*-Toludine (3·8 g)	*m*-Tolyl guanidine[5]	65	215–218 (Sulphate)
4.	*p*-Toludine (3·8 g)	*p*-Tolylguanidine[5]	92	163 (Carbonate)
5.	*o*-Chloro aniline (4·2 g)	*o*-Chlorophenylguanidine[6]	90	154 (Picrate)
6.	*m*-Chloro aniline (4·2 g)	*m*-Chlorophenylguanidine[6]	90	160 (Picrate)
7.	*p*-Chloro aniline (4·2 g)	*p*-Chlorophenylguanidine[6]	90	154 (Carbonate)
8.	*o*-Anisiidne (4·1 g)	*o*-Anisylguanidine[7]	85	150 (Picrate)
9.	*m*-Anisidine (4·1 g)	*m*-Anisylguanidine[7]	85	145 (Picrate)
10.	*p*-Anisidine (4·1 g)	*p*-Anisylguanidine[7]	85	130 (Carbonate)
11.	*o*-Phenetidine (4·5 g)	*o*-Phenetylguanidine[8]	80	160 (Carbonate)
12.	*p*-Phenetidine (4·5 g)	*p*-Phenetylguanidine[8]	80	165 (Carbonate)
13.	2, 5-Dimethoxy-aniline (5·1 g)	2, 5-Dimethoxyphenyl-guanidine[5]	75	148 (Picrate)
14.	2, 5-Diethoxy aniline (5·8 g)	2, 5-Diethoxyphenyl guanidine[8]	70	140 (Picrate)
15.	2, 5-Dichloro aniline (5·3 g)	2, 5-Dichlorophenyl guanidine[6]	65	165 (Picrate)
16.	Benzylamine (3·6 g)	Benzylguanidine[2,6]	70	185 (Picrate)
17.	Methylamine (1·0 g)	Methylguanidine[2,6]	40	170 (Hydrochloride)
18.	Ethylamine (1·5 g)	Ethylguanidine[2,6]	45	175 (Picrate)
19.	*n*-Butylamine (2·4 g)	*n*-Butylguanidine[2,6]	70	178 (Picrate)

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---

1. Rose, F. L. and Swain, G., *J. Chem. Soc.*, 1966, **44**, 22.
2. Singh, G. C., *Jour. Indian Chem. Soc.*, 1968, **45**, 27.
3. (*a*) Claus, A., *Ann.*, 1875, **179**, 139.
   (*b*) Werner, E. A., *J. Chem. Soc.*, 1912, **101**, 2166.
4. Kampf, Adolf, *Ber.*, 1904, **37**, 1683.
5. Braun, C. E., *J. Amer. Chem. Soc.*, 1933, **55**, 1281.
6. Smith, G. B. L., *Ibid.*, 1929, **51**, 477.
7. Wheeler, R. C. and Johnson, M. A., *Am. Chem. Journ.*, 1901, **26**, 417.
8. Davis, T. L. and Rosenquist, E. N., *J. Amer. Chem. Soc.*, 1937, **59**, 2112.

EFFECT OF STAGE OF CUTTING ON THE CRUDE PROTEIN, Ca AND P CONTENT OF LUCERNE (*MEDICAGO SATIVA* L.) VARIETIES

LUCERNE is one of the most palatable and nutritious food of the forage legumes. In this study the crude protein, phosphorus and calcium contents of different varieties of lucerne were studied at monthly intervals.

Fifteen cultivars of lucerne were grown during 1974–75. The first cut was made after the second month and the subsequent cuttings at monthly intervals. The plant samples obtained from eight cuts were analysed for crude protein[2] (N% × 6·25), Ca and P[3]. The mean values for crude protein, calcium, phosphorus for five important varieties are given in Table I.

TABLE I

Variety	Crude protein %	Calcium %	Phosphorus %
Anand-1	23·2	2·46	0·22
Anand-2	21·6	2·36	0·22
Sirsa-9	22·8	2·10	0·23
Hissar	22·8	2·18	0·22
Coimbatore	21·3	2·28	0·23

TABLE II

Month	2	3	4	5	6	7	8	9	C.D. at 5%
Crude protein %	21·5	23·9	24·3	23·3	23·2	23·4	17·6	20·6	1·52
Calcium %	2·34	2·31	2·26	2·67	2·45	2·05	1·90	1·80	0·18
Phosphorus %	0·20	0·22	0·25	0·22	0·25	0·25	0·18	0·17	0·019

The different varieties did not differ in their protein and phosphorus contents. However they differed in their Ca content (C. D. = 0·24 at 5%).

The mean values of fifteen lucerne varieties for crude protein, calcium and phosphorus content on dry weight basis at different cutting intervals are presented in Table II.

All the three constituents differed significantly with the cutting intervals. The maximum crude protein was recorded in the cutting taken at fourth month followed by that of the third month. The crude protein content decreased from the fourth month onwards. Earlier investigators[1,4] observed a general decrease in the protein content with the maturity of the crop but the period of maximum concentration of crude protein was not reported. The calcium and phosphorus contents were maximum during fifth and the fourth months respectively and decreased later on[5]. It could therefore be concluded that the lucerne varieties had the maximum concentration of crude protein, calcium and phosphorus in the fourth and fifth month cuttings.

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---

1. Gonzalez, G. and Trevino, J., *Herbage Abstr.*, 1975, **45**, 18.
2. Humphries, E. C., *Modern Methods of Plant Analysis*, Springer-Verlag, Berlin, 1956, **1**, 468.
3. Jackson, M. L., *Soil Chemical Analysis*, Prentice Hall of India (Pvt.) Ltd., New Delhi, 1973.
4. Lee, C. T. and Smith, D., *Agron. J.*, 1972, **64**, 326.
5. Polidori, F., D'urso, G., Quila, D. A. S. and Sarro, F., *Herbage Abstr.*, 1973, **43**, 145.

SOME PARASITES OF CABBAGE WORM, *PIERIS BRASSICAE* LINN. (LEPIDOPTERA : PIERIDAE) FROM (KULU VALLEY), HIMACHAL PRADESH

THE cabbage worm, *Pieris brassicae* Linn., is a serious pest of cruciferous crops and is widely distributed in many parts of the world. In India, it causes appreciable damage in Punjab, Himachal Pradesh, Uttar Pradesh, Bihar, Bengal and Assam. At Vegetable Research Station, Katrain, Kulu Valley, a collection of eggs, larvae and pupae of *P. brassicae* was made during different months from 1972 to 1974 and following parasites were recovered in the laboratory.

No egg parasite was found. Among the larval parasites, *Apanteles glomeratus* L. was very common, attacking about 13% to 25% of the larvae in May and about 30% in June. *Tetrastichus sokolowskii* Kurdj. is a new record on *P. brassicae*. But, this parasite has been previously recorded on diamond back moth *Plutella xylostella* L.[1,2,10] from America and India. Its parasitization during May and June was between 7% and 10%. The percentage parasitization of *Apanteles* sp., *Ecphoropsis perdistinctus* Vier and *Bracon* sp. was 4%, 1·3% and 2·5%, respectively.

TABLE I

Parasites recovered in the laboratory on the eggs, larvae and pupae of Pieris brassicae *Linn.*

Order/Family	Parasites	Stage attacked	Parasitism (%)
Hymenoptera			
Braconidae	1. *Apanteles* sp.	Caterpillar	4
	2. *A. glomeratus* L.	,,	13–30
	3. *Bracon* sp.	,	2·5
Chalcididae	4. *Brachymeria* sp.	Pupa	2·3
Eulophidae	5. *Tetrastichus sokolowskii* Kurdj.	Caterpillar	7–10
Ichneumonidae	6. *Nythobia* (*Diadegma*) sp.	Pupa	2-4·5
	7. *Ecphoropsis perdistinctus* Vier.	Caterpillar	1·3
Pteromalidae	8. *Pteromalus puparum* L.	Pupa	20–35

The pupal parasite, *Pteromalus puparum* L., was the most common and it attacked about 20% to 35% pupae. The other two pupal parasites, *viz.*, *Nythobia* (*Diadegma*) sp. and *Brachymeria* sp. are the first records on *P. brassicae* from India. Some species of the genus *Nythobia* (*Diadegma*) have been recorded on other crop pests[3,5,7-9]. *Nythobia* (*Diadegma*) sp. is a black coloured ichneumonid parasite. The front and middle leg segments are light yellow, hind coxae and tarsi black and rest of the leg light yellow. It is about 4 mm in length and 0·9 mm in width across the head. The parasitization by *Nythobia* (*Diadegma*) sp. was only about 2% to 4·5%. Some of the species of *Brachymeria* have been recorded on other crop pests, *e.g.*, *Brachymeria bicolorata* on *Earias* sp. in Pakistan[4], *Brachymeria phya* on *Plutella xylostella* L. in Canada[11], *Brachymeria* sp. on *Plutella xylostella* L. in India[6] and *Brachymeria femorata* on *Pieris brassicae* L. in Bulgaria[3]. Its parasitism was found at 2·3% only.

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---

1. Cherian, M. C. and Basheer, M., *Proc. Indian Acad. Sci.*, 1939, **1 B**, 87.
2. Harcourt, D. G., *Canad. Ent.*, 1960, **94**, 419.
3. Kaitazov, A., *Rast. Zasht.*, 1963, **11**, 20.
4. Khokhar, K. F., Qudri, M. A. H. and Ahmed, M., *Pakistan J. Sci. Ind. Res.*, 1971, **14**, 261.
5. Ootmann, E. R. and Plantner, G. R., *Hilgardia*, 1969, **40**, 1.
6. Patel, V. C. and Patel, H. K., *Indian J. Ent.*, 1968, **30**, 86.
7. Rao, V. P. and Nair, K. R., *Tech. Bull. Commonwealth Inst. Biol. Control*, 1967, **9**, 73.
8. Rojas, P. S., *Agriculture Tec.*, 1965, **25**, 39.
9. Tikar, D. T. and Thakre, K. R., *Indian J. Ent.*, 1962, **23**, 116.
10. Ullyett, G. C., *Ent. Mem-Dep. Agric. S. Afr.*, 1947, **2**, 77.
11. Yarrow, W. H. T., *Queen. J. Agril. An. Sci.*, 1970, **27**, 321.

A NOTE ON THE CYTOLOGY OF *BOERHAAVIA REPANDA* WILLD.

THE present study deals with the karyomorphology and meiosis of *B. repanda* Willd. The material was collected locally and grown in pots. For somatic preparations, excised root tips were pretreated with 8-hydroxyquinoline for about 2½ hrs at 10° to 15° C, and stained according to Feulgen's schedule. Permanent slides were made by passing through acetic acid butanol series and mounted in euparal. Meiotic studies were made by fixing the flower buds at different stages of development in 1 : 3 acetic alcohol for 24 hrs. Smear preparations were made in 1% propionocarmine.

Karyotype description is made following Levan *et al.*[2]. For determining the type of chromosome the centromeric index ($r = l/s$), where 'r' as the arm ratio, 'l' as the length of the long arm, and 's' as the length of the short arm is taken into account. The diploid chromosome number of this plant is found to be $2n = 42$ (Figs. 1, 2). The chromosomes fall in a series of close gradations. However, a pair of long submedian chromosomes ($2 \cdot 36\,\mu$) is distinguishable in the karyotype. There are five pairs of chromosomes with median centromeres, and all the rest of the chromosomes show submedian centromeres. The chromosome-measurements and classification are given in Table I.

TABLE I

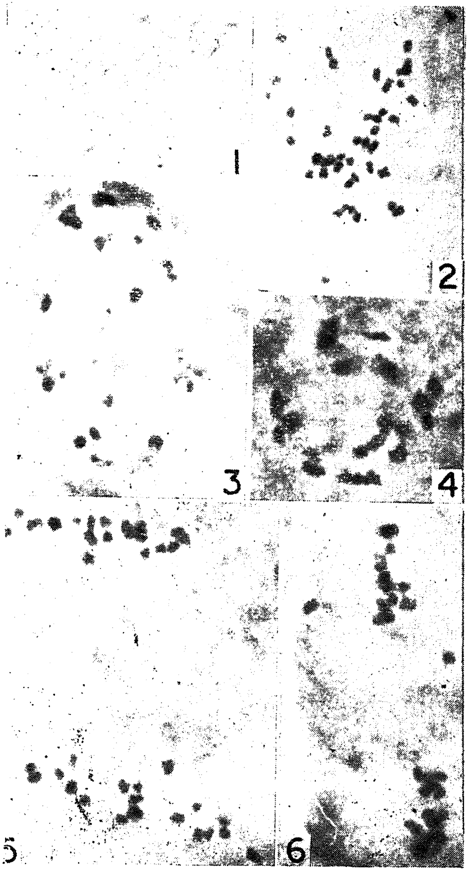
Karyotype analysis in B. repanda *Willd.*

Chromosomes	Length in microns: Long arm	Length in microns: Short arm	Total length in μ	Arm ratio	Centromere
I	1·62	0·74	2·36	2·18	Sm
II	1·21	1·08	2·29	1·12	Sm
III	1·08	0·74	1·82	1·45	Sm
IV	1·14	0·81	1·95	1·53	Sm
V	1·08	0·81	1·89	1·33	Sm
VI	1·08	0·81	1·89	1·33	Sm
VII	1·08	0·81	1·89	1·33	Sm
VIII	1·08	0·81	1·89	1·33	Sm
IX	1·02	0·81	1·83	1·14	Sm
X	1·08	0·74	1·82	1·45	Sm
XI	0·88	0·88	1·76	1·00	M
XII	0·94	0·74	1·68	1·27	Sm
XIII	0·88	0·81	1·69	1·09	Sm
XIV	0·81	0·81	1·62	1·00	M
XV	0·81	0·81	1·62	1·00	M
XVI	0·81	0·81	1·62	1·00	M
XVII	0·81	0·74	1·55	1·09	Sm
XVIII	0·81	0·54	1·35	1·50	Sm
XIX	0·67	0·60	1·27	1·11	Sm
XX	0·67	0·54	1·21	1·25	Sm
XXI	0·54	0·54	1·08	1·00	M

Meiosis is found to be mostly regular. At diakinesis and metaphase-I 21 bivalents are observed (Figs. 3, 4). A bivalent is attached to the nucleolus at diakinesis (Fig. 3). There is regular disjunction of chromosomes at anaphase-I (Fig. 5). However, occasionally one or two precociously moving chromosomes are observed at metaphase-II (Fig. 6).

The chromosome number reported for *B. diffusa*[6,3,1,4] as $n = 58, 47, 13$ and $2n = 26$ is quite varying. For *B. repanda* the only report is $n = 21$ by Tandon and Rao[5]. It was the intention to know whether the chromosome number in *B. repanda* is also verying or not. The present studies both of somatic and meiotic chromosomes confirm the findings of Tandon and Rao[5] in that the chromosome number in *B. repanda* was not varying as in *B. diffusa*.

The author is grateful to Prof. M. S. Chennaveeraiah, Head of the Department of Botany, Karnatak University, Dharwar, for encouragement and valuable suggestions.



FIGS. 1–6. Figs. 1–2. Photomicrograph of somatic metaphase chromosomes ($2n = 42$), × 1,800. Figs. 3–6. Meiosis, × 1,000. Fig. 3. Diakinesis showing 21 bivalents. Fig. 4. Metaphase-I chromosomes showing 21 bivalents. Fig. 5. Anaphase-I showing equal distribution of chromosomes. Fig. 6 Metaphase-II showing 2 precociously moving chromosomes.

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Dharwar-3, *January* 31, 1976.

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1. Gill, L. S. and Abubakar, A. M., "IORB Chromosome Number Report," *Taxon*, 1975, 24, 371.
2. Levan, A., Fredga, K. and Sandberg, A. A., *Hereditas*, 1964, 52, 201.
3. Sharma, A. K. and Bhattacharyya, G. C., *Ind. Agriculturist*, 1961, 5, 9.
4. Srivastava, A. K. and Misra, K. C., *Science and Culture*, 1966, 32, 315.
5. Tandon, S. L. and Rao, G. R., *Curr. Sci.*, 1963, 32, 234.
6. Thombre, M. V., *Science and Culture*, 1959, 25, 208.

A NEW LEAF-SPOT DISEASE OF RAMIE IN INDIA

In the course of investigations on the diseases of ramie (*Boehmeria nivea*) a leaf-spot disease caused by *Myrothecium roridum* Tode ex. Fr. has been observed at the Ramie Research Station, Sorbhog, Assam. This incidence has also been noted on ramie at Debi jhora area of West Dinajpur in West Bengal. There are reports in India that *M. roridum* attacks various plants of agricultural importance and causes considerable damage to them[1-4].

The first symptom appears as irregular, small, round, tan coloured spots about one mm. in diameter on the upper surface of the lamina. As the disease advances the spots become circular to irregular, 6–16 mm. in diameter and brown to dark brown in colour. In cases of severity the incidence of the disease is as high as 7–10%. On maturity the sporodochia develop in concentric rings as black dot like structures in the central regions of the spots.

Ramie plants (24 days old) were inoculated by spraying the spore suspension. Three days after inoculation similar symptoms appeared on the leaves. Fungal fructification was, however, noted after six days of inoculation.

On the basis of the morphological characters the fungus has been identified as *Myrothecium roridum* Tode ex. Fr. Since there is no report of this fungus on ramie the present investigation thus appears to be the first record form India or elsewhere.

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1. Munial, R. L., *Indian Phytopath.*, 1960, **13**, 150.
2. Mukherjee, N. and Basak, M., *Indian J. Mycol. Res.*, 1969, 7 (1), 37.
3. Saksena, H. K. and Tripathi, R. C., *Indian J. Mycol. and Pl. Path.*, 1971, 1 (1), 75.
4. Singh, D. V. and Srivastava, H. S., *Indian J. Miocrobiol.*, 1967, 7, 39.

NUTRITIONAL VALUE OF *MORINGA*

Moringa or drumstick, [*M. oleifera* ; *M. concanensis*] locally called 'saijan', is well known as a poor man feed in so far as its leaves and pods are concerned. The pods and leaves have been reported[1] to be a rich source of vitamin C. *Moringa* attained significance in the last few years in view of its wood being a potential source of raw material for paper industry ; more so because it is a fast growing tree. A large-scale cultivation of *Moringa* is underway to meet the paper shortage. With this end in view a survey of *Moringa* plants was made in Lucknow District and 11 clones of *M. oleifera* and 2 clones of *M. concanensis* were selected and protein and fat were estimated at species level (*M. oleifera* and *M. concanensis*) whereas the vitamin C content was estimated at clonal level.

The seeds obtained from the mature fruits of *M. oleifera* and *M. concanensis* after extraction with *n*-hexane yielded pale yellow fat 27% and 33% respectively. The physical constants of *M. oleifera* fat were found to be, $n_D^{25°}$, 1·46 ; acid value, 0·26 ; iodine value, 66·1 ; while those of *M. concanensis* were $n_D^{25°}$, 1·46 ; acid value, 0·25 and iodine value. 67·07. The fatty acid composition of the seed fat of the two species by GLC revealed that the fats are rich in oleic acid and the composition of *M. oleifera* fat is in contradistinction to the observation of Rao *et al.*[2] (Table I).

Table I

Fatty acid composition

Fatty Acids	Present investigation *M. oleifera* %	Present investigation *M. concanensis* %	Rao *et al.* *M. oleifera* %
Lauric	12·5	0·6	..
Palmitic	9·6	9·1	9·3
Palmito-oleic	1·6	2·8	..
Stearic	3·0	2·4	7·4
Behenic	..	..	8·6
Oleic	73·3	83·8	65·7

The unsaponifiable portion (0·05%) of the seed fat from both the species on purification yielded small amounts of a sterol which on co-argentation TLC (silica gel G ; petroleum ether-chloroform-acetic acid, 75 : 25 : 2 ; 10% H_2SO_4 (v/v) spray) corresponded to β-sitosterol.

The detoxified seed meals of *M. oleifera* and *M. concanensis* were found to be rich in protein, 50% and 72·6% respectively. The seed meals are being tried as poultry feed and can also be considered for microbial fermentation.

The moisture and vitamin C contents of the fresh leaves as well as pods of various clones (11 varieties of *M. oleifera* and 2 varieties of *M. concanensis*) at two different stages, (*i*) tender and soft, (*ii*) mature and hard were determined[3] (Table II).

TABLE II

Moisture and vitamin C contents in leaves and pods

Clones	Moisture percentage			Vitamin C in mg/100 g		
	Leaves	Pods tender	Pods mature	Leaves	Pods tender	Pods mature
M.O. 2	82·58	83·14	85·98	86·36	91·09	103·19
M.O. 2A	77·87	84·79	85·91	105·00	75·76	93·01
M.O. 3	84·03	83·21	84·66	79·06	99·34	121·38
M.O. 5A	87·61	87·10	86·72	94·84	96·69	112·58
M.O. 5B	83·69	83·47	88·95	95·53	103·69	136·47
M.O. 7	81·84	82·83	85·17	105·26	105·05	128·87
M.O. 10	81·06	85·09	85·29	107·20	91·34	112·57
M.O. 11	71·90	84·58	85·92	113·39	99·73	109·84
M.O. 12	71·69	84·65	86·69	133·64	105·58	122·37
M.O. 13	78·58	86·57	83·92	80·35	131·64	142·58
M.O. 16	76·04	82·66	83·81	99·41	113·51	143·87
M.C. 1	82·77	82·95	86·22	59·00	106·67	130·05
M.C. 3	80·25	86·65	87·78	55·01	75·74	103·49

M.O. = *Moringa oleifera*, M.C. = *Moringa concanensis*.

The moisture in the leaves of *M. oleifera* ranged from 71–84% and in that of *M. concanensis* from 80–82% and the vitamin C content (mg/100 g) was 79–133 in former and 55–59 in the latter species. The moisture content at tender and mature stages of the pods of *M. oleifera* ranged from 82–87% and 83–88% respectively, while in those of *M. concanensis* it ranged from 82–86% and 86–87%. The vitamin C content in tender stage of the pods of different clones of *M. oleifera* varied from 75–131 and in mature stage it varied from 93–143, whereas in those of *M. concanensis* it varied from 75–106 and 103–130 respectively.

From the results it can be concluded that the seeds of *M. concanensis* yield higher percentage of protein and fat, richer in oleic acid as compared to *M. oleifera* (Table I). So far as the vitamin C content is concerned, *M. oleifera* leaves and pods are a better source than those of *M. concanensis* (Table II). *Moringa* thus have the potentiality of the newer source of fat, protein and vitamin C.

Thanks are due to Dr. P. D. Dogra for kindly authenticating the plant material and to C.S.I.R., for the award of Senior Research Fellowship to R. Banerji.

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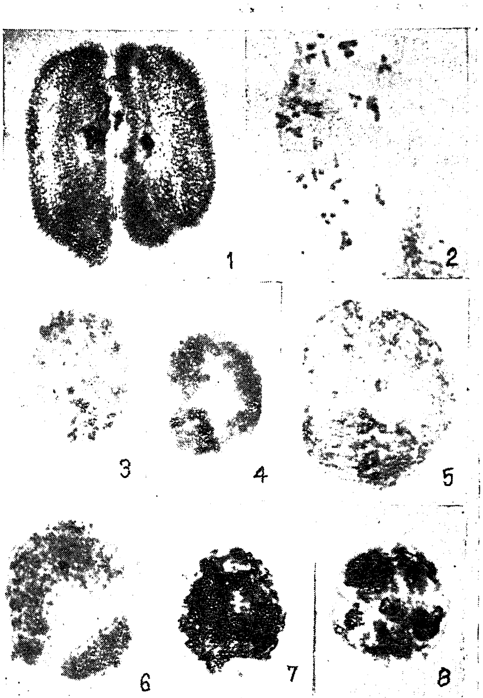
1. Dogra, P. D., Singh, B. P. and Tandon, S., *Curr. Sci.*, 1975, **44**, 31.
2. Rao Subba, B. C, Rao Narayana K. and and Jois, Subba H., *J. Indian Chem. Soc.*, 1953, **30**, 477.
3. Glik, E. O., *Methods of Biochemical Analysis*, Vol, I, p. 127.

POLARITY OF METAPHASE PLATE IN *DIOSCOREA HISPIDA* DENST.

D. hispida is a climber with prickly stem twinning to the left with trifoliate hairy leaves and closely resembles that of the African *D. dumetorum*. In the male plants, the flowers do not open and they remain in the closed condition and later the inflorescences slowly wither away. The male flowers contain only shrunken anthers devoid of pollen grains. Mostly the anther lobes are in a collapsed condition (Fig. 1).

To ascertain the cause of male sterility, the microsporogenesis was studied in detail. Early development of pollen mother cells and tapetum were normal. The chromosome number was determined as $2n = 60$. Trivalents, bivalents and univalents have been noted (Fig. 2). Though the development of sporocytes was normal upto M-1, later stages were characteristically absent.

At early M-I the chromosomes, in variable association, remained scattered throughout the cell. However, gradually all the chromosomes were found at the periphery on one side of the cell (Fig. 3). Later the chromosomes formed an equatorial plate. Frequently, the metaphase plates were oriented parallel to the long axis of the PMCs instead of orienting at 90° to the long axis when they were not globular in shape. There was complete failure of anaphase and in the meantime cytokinesis occurred (Fig. 4).



FIGS. 1–8. Fig. 1. Empty anther lobes, × 55. Fig. 2. M-I with trivalents, bivalents and univalents ($2n = 60$), × 2,200. Fig. 3. Chromosome pushed towards periphery, × 900. Fig. 4. Disturbed polarity of metaphase plate, × 900. Fig. 5. Disturbed polarity of metaphase plate and spindle, × 1,400. Fig. 6. Unequal dyad cells, × 1,400. Fig. 1. Degenerating PMC, × 900. Fig. 8. Degenerating PMC with micronuclei, × 900.

However, in a few cells spindles were organised in the plane of short axis, thereby ruling out the possibility of distribution of chromosomes in a normal way between the cells. In the meantime wall formation took place resulting in a dyad of unequal size (Figs. 5, 6). In more than 90% of pollen mother cells the entire metaphase plate was included in the smaller dyad cell. Nevertheless, occasionally, the wall formation may take place in such a way to divide the metaphase plate unequally among the dyad cells. Due to failure of cytokinesis, a few cells degenerated *en masse* (Fig. 7). In some cases micronuclei are organized leading to the degeneration of PMCs (Fig. 8). Further, the tapetum also showed signs of degeneration ultimately rendering the anther sacs empty.

The bivalents are invariably found oriented on their long axis of the spindle, instead of congressing at the equatorial plate as in mitosis[6]. Though suppression of spindle formation in *C. mitosis* and the development of abnormal spindles are well known[1,4,7], the occurrence of metaphase plate with disturbed polarity is reported for the first time in plants.

Since *Dioscorea* is a vegetatively propagated crop, a high degree of male and female sterility prevails. This is mainly attributed to the presence of polyploid series in the genus. The present study shows that the mechanism of male sterility operating in *Dioscorea* may be of different types. Though various types of male sterility have been reported in different plants[2,3,5], the occurrence of male sterility due to the disturbed orientation of metaphase plate followed by failure of anaphase movement in *Dioscorea* is rather interesting. Further studies may reveal the extent to which the present mechanism is responsible for the manifestation of sterility in other species.

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---

1. Deysson, G., *Int. Rev. Cytol.*, 1968, 24, 99.
2. Jain, S. K., *Biblio. Genetica,* 1959, 18, 101.
3. Jos, J. S. and Magoon, M. L., *Ind. Jour. Hort.*, 1971, 28, 224.
4. Kabarity, A., *Cytologia,* 1966, 31, 457.
5. Magoon, M. L. and Jos, J. S., *The Nucleus,* 1968, 11, 1.
6. Swanson, C. P., *Cytology and Cytogenetics,* MacMillan and Co. Ltd., London, 1963.
7. Vaarama, A., *Hereditas,* 1949, 35, 136.

X-RAY STUDIES ON THE EFFECT OF *BACILLUS THURINGIENSIS* BERLINER ON THE FEEDING ACTIVITY IN THREE SPECIES OF LEPIDOPTERA

THE effect of *Bacillus thuringiensis* Berliner has been studied on the citrus leaf caterpillar, *Papilio demoleus* L.[1], the castor semi-looper, *Achoea janata* L.[2] and the castor leaf caterpillar, *Spodoptera litura* (F.)[3]. While the former two species showed symptoms of general paralysis and gut paralysis respectively, the latter was found to be not susceptible to the pathogen. It has been shown that the susceptible species do not feed again after an initial meal on contaminated foliage with the crystal-forming bacterium, *B. thuringiensis*, thus extensive defoliation during the incubation period of the disease is prevented. In the present study, observations on the bowel movement in the above three insects after treatment with *B. thuringiensis* have been made by taking X-ray photographs.

Finely powdered barium sulphate was mixed with sterile water to form a thick paste. This was added to an equal volume of Biotrol wettable powder containing 30 billion viable spores of *B. thuringiensis* per gram, and the mixture was painted on fresh foliage of the respcetive host plant, air dried and fed to test insects. The larvae were starved for 12 hr. before the commencement of the experiment. The treated larvae were taken 8 hr. after the feeding, and washed in water thoroughly to remove the adherent particles of $BaSO_4$. The larvae were placed on Agfa Normal film and the X-ray photographs were taken by using Siemens-Reinigerwerke X-ray machine with an exposure of 2 seconds at 3 ma and 50 kV. The excreta collected from the treated larvae were also X-ray photographed.

It was observed that the food travelled the entire region of the alimentary canal without any obstruction in healthy *P. demoleus, A. janata* and *S. litura.* The presence of $BaSO_4$ was also confirmed in their excreta. In the case of *P. demoleus*, treated with $BaSO_4$ and *B. thuringiensis*, feeding has been stopped within 8 hr since food has remained in the foregut itself due to stoppage of bowel movement. The histopathological studies made earlier also showed complete breakdown of gut epithelium[1]. In the case of *A. janata* also there was a blocking of bowel movement in the alimentary canal but food passed through up to the midgut. The histopathological studies made on this insect revealed the severity of gut damage[2]. On the other hand, in the case of *S. litura,* the X-ray study showed no stoppage of food material in the alimentary canal. The faecal pellets collected from this insect 12 hr after feeding also revealed the presence of $BaSO_4$, thus confirming the food movement through the alimentary canal without any obstruction. This is an additional evidence for the non-susceptibility of this insect to *B. thuringiensis* reported by Govindarajan *et al.*[3].

It is now realised, however, that the symptoms of general paralysis caused by crystalliferous bacteria is the exception rather than the rule, with reference to most of the susceptible lepidoptera[4]. Based on the X-ray photographic study and other histological and blood and gut pH studies, the above three species of insects may be grouped under type I, type II and type IV category showing general paralysis, gut paralysis and non-susceptible categories respectively as suggested by Angus and Heimpel[5] and Martouret[6].

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---

1. Narayanan, K. and Jayaraj, S., *Indian J. exp. Biol.*, 1974, 12, 89.
2. Govindarajan, R., Jayaraj, S. and Narayanan, K., *Z. ang. Ent.*, 1976 (in Press).
3. —, — and —, *Indian J. exp. Biol.*, 1975, 13, 548.
4. Heimpel, A. M. and Angus, T. A., *J. Insect Pathol.*, 1959, 1, 152.
5. Angus, T. A. and Heimpel, A. M., *Canad. Ent.*, 1959, 91, 352.
6. Martouret, D., *Symp. Phytopharm. Phytiat.*, 12th, Ghent, Belgium, 1961, 8, 1.

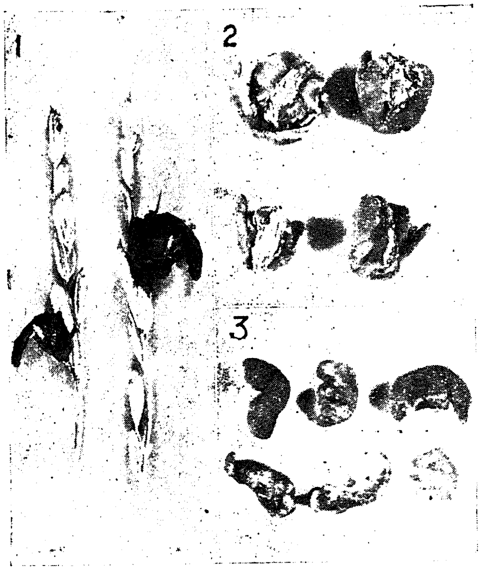
OCCURRENCE OF TRUE SCLEROTIA IN *CLAVICEPS ORYZAE-SATIVAE*—THE CAUSAL ORGANISM OF FALSE SMUT OF RICE

'FALSE SMUT' of rice caused by *Claviceps oryzae-sativae* Hashioka [imperfect stage *Ustilaginoidea virens* (Cke.) Tak.] is considered to be a minor disease, but its occurrence has been reported from almost all the rice-growing countries in Far-East, South-East Asia, Africa, North and South America. However, under favourable conditions, the disease causes considerable loss in grain yield. In India, the disease is common in coastal parts of Orissa, Andhra Pradesh, Haryana and Tarai and Hills of Uttar Pradesh. Of late, the incidence of the disease is reported to be on an increase especially on the high yielding varieties.

The symptom of the disease becomes discernible after the flowering when the ovary of the infected kernel is transformed into a large velvety, yellow to orange pulverulent mass (pesudomorph) changing to olive green in colour. The pseudomorph covered with spore-mass had been erroneously termed as "sclerotia" by earlier workers which never produced ascigerous state of the fungus. The formation of true sclerotium, its germination and formation of perithecia and ascospores have been demonstrated in Japan[1-3,5] but has not been reported from any other country.

Extensive surveys for the incidence of this disease in Uttar Pradesh and Haryana and the careful examination of the several specimens received from Andhra Pradesh, Orissa and Bihar revealed that the true sclerotia of the fungus are not produced in the plains. The formation of true sclerotia was observed in almost all the collections done at an altitude of 1200 m and above in Kumaon region of Uttar Pradesh.

After the dispersal of conidia, true sclerotia remained loosely attached on the pseudomorphs and may fall in the field with slight jerk (Fig. 1).



FIGS. 1–3. False smut of rice. Fig. 1. Spikelets showing true sclerotia hanging on the pseudomorph, × 1·2. Fig. 2. True sclerotia covering the spore ball, × 1·75. Fig. 3. True sclerotia, × 2.

The number of true sclerotia per ball varies from 1 to 5 but 2 are most frequently observed. These sclerotia remained attached separately or sometimes joined to cover the pseudomorph like an umbrella (Fig. 2). The sclerotia are hard, dark-brown, clavate, botuliform, reniform, horse-shoe shaped to indefinite in shape, concave on the inner-side and convex on the outer side measuring 3–10 mm × 1–5 mm and 0·5–1–5 mm. in thickness (Fig. 3). The transverse section of the sclerotia shows the outer tissue to be made of compact pseudoparenchymatous cells while the inner tissue is hyaline with prosenchymatous cells. Ou[4] mentions that some of the green spore balls develop 1 to 4 sclerotia in the centre. Hundreds of green balls, especially bigger ones were examined for true sclerotia but could not be observed in the centre.

This constitutes the first report for the occurrence of the true sclerotia in the country. Further work on its germination and role in the disease cycle are in progress.

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1. Hashioka, Y., *Il. Riso,* 1971, 30, 235.
2. —, Yoshino, M. and Yamamoto, T., *Saitama Agr. Expt. Sta. Res. Bull.,* 1951, 2, 1.
3. Hemni, T. and Konishi, Z., *J. Pl. Prot.*, 1939, 26, 857.
4. Ou, S. H., *Rice Diseases,* Commonwealth Mycological Institute, Kew, 1972, p. 368.
5. Sakurari, M., *Ann. Phytopath. Soc. Jap.,* 1934, p. 70.

EMS-INDUCED MULTICARPELLATE CONDITION IN *CAJANUS CAJAN*

DURING the course of the observations on M_1 progenies of pigeon pea treated with EMS, a number of morphological variants were observed under different treatments. The most interesting among them which we wish to report here is the multicarpellate condition observed in an early maturing variety, treated with 0·03 M. EMS.

Although the plant produced abundant flowers, it was found to be devoid of fruits at the time of harvest. A closer examination of several flowers of the plant revealed a tendency for the presence of supernumerary petals (8 to 10 instead of 5). Some of the flowers showed 12 stamens (11) + 1 instead of 10. Some of the stamens also appeared to be transformed into sepal-like or petal-like structures.

Examination of different flowers of that plant showed the following three types of pistils (Fig. 1).

(1) Normal sized flower with one carpel.

(2) Slightly bigger flower having bicarpellary apocarpous condition.

(3) Biggest flowers with 8–10 petals, 10–12 stamens and tricarpellary apocarpous condition (Fig. 2).

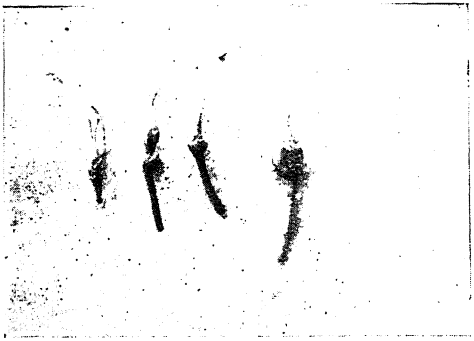

FIG. 1. Intra-plant variation in Carpel number, excluding the longest and rightmost carpel which represents the control.

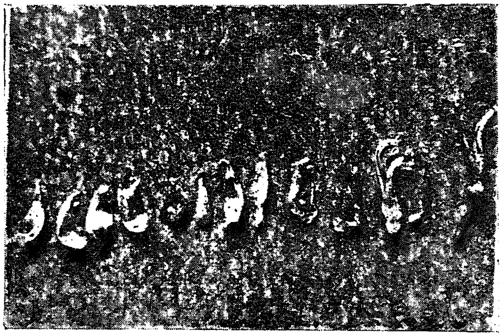

FIG. 2. A tricarpellate flower with 12 (11 + 1) stamens and 10 petals.

All the three types of flowers were both pollen and ovule sterile and failed to set seeds. Though the short gynophores of the apocarpous ovaries were fused, the carpels remained free. In addition, many of the ovaries in the second and the third case were found to have a longitudinal slit through which the ovules were exposed. The plant looks abnormal having lower height and less number of branches.

In the family leguminosae, bicarpellary and apocarpous condition was reported in *Crotalaria laburmifolia* by Rao[5], in *Vigna sinensis* by Dutta[1] and Henry Louis[2], and in *Cajanus cajan* by Joshi *et al.*[3]. An obcardate mutant with polycarpellary and apocarpous condition (with two to three pods in each calyx) was reported in pigeon pea by Kajjari[4].

The present variant however, differs from the multicarpellary mutant of Joshi *et al.*, and obcardate mutant of Kajjari in having all the three types of flowers (*i.e.*, mono-, bi- and tri-carpellary) on the same plant. It also differs from their mutants by having complete pollen and ovule sterility.

Though multicarpellate condition has already been reported in this crop, the present observation appears to be the first instance where all the three types of flowers (mono-, bi- and tri-carpellary) are found on the same plant with concomitant increase in the stamens and complete pollen and ovule sterility.

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Faculty of Agriculture,
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Varanasi, *February* 6, 1976.

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R. M. SINGH.
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---

1. Dutta, T. R., *Sci. and Cult.*, 1956, 22, 29.
2. Henry Louis and Kadambasundaram, M., *Madras Agri. J.*, 1963, 60, 9.
3. Joshi, B. C. and Ramanujam, S., *Indian J. Genet.*, 1963, 23, 64.
4. Kajjari, N. B., *Curr. Sci.*, 1956, 25, 333.
5. Rao, K. V. and Sambasiva, *Ibid.*, 1954, 20, 416.

SHORT SCIENTIFIC NOTES

Telenomus manolus Nixon, A Potential Egg Parasite of Groundnut Red Hairy Caterpillar, *Amsacta albistriga* Walker

The Red hairy caterpillar is a pernicious menace to the groundnut crop in India. The crop raised in dry land is more prone to the pest than the crop in irrigated soils. A search for the natural enemies associated with the pest was made in Tamil Nadu from August to October, 1974. The results of the study are presented in this paper.

The parasites emerged from the eggs belonged to the species, *Telenomus menolus*[1] which has not so far been reported to parasitise the eggs of *A. albistriga* either in India or elsewhere[2].

The results also indicate that the degree of parasitisation was as high as 94·3% with 3·8 to 59·4% emergence of adult (Table I). The poor emergence might be due to multiparasitism or due to existing high temperature ranging from 37° to 40° C. The high degree of parasitisation observed, suggests that this could be effectively used along with other methods to control this pest.

TABLE I

Degree of parasitisation and per cent emergence of Telenomus manolus *in different localities*

Name of locality	No. of eggs observed	Per cent parasitisation	Per cent emergence of parasites
Kuttupatty	307	76·2	59·4
Sanarpatty	163	83·4	41·2
Natham	268	90·6	20·6
Kappalur	236	84·7	21·5
Sekkanoorani	283	94·3	3·8

The authors are thankful to Dr. R. W. Fennah, Director, British Museum, Commonwealth Institute of Entomology, London, for his kind help in identifying the parasite and providing informations on its distribution.

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University,
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January 21, 1976.

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1. Fennah, R. G., Personal Communication, 1974.
2. —, Personal Communication, 1975.

Larval Colour Mutants of *Culex fatigans*

Interest in formal genetics and cytogenetics of mosquitoes has grown considerably during recent years. The development of resistance to chemicals and the unequivocal demonstration of its genetic basis have underlined the need for extensive genetic studies of mosquitoes, especially of the vector species. This interest in mosquito genetics has been sustained mainly by the continuing discovery of new mutant forms. At present a few mutants are known in the genus *Culex*. Laven's review (1967)[1] of the Genetics, speciation and Evolution of *Culex pipiens* is a distinct landmark.

The seven laboratory strains of *Culex fatigans*, the chief vector of filariasis in the tropics, used in the present studies have given ample evidence of the existence of extensive genetic variability (Shetty, 1974)[2]. Five larval colour mutants of *Culex fatigans* have been established. These mutants are golden yellow (*go*), green (*g*), brown (*br*), greyish brown (*gb*) and dark (*da*). The mechanism of inheritance of these mutants was found to be controlled by a single autosomal recessive factor. Crossing experiments between the wild-type and these mutants gave a perfect 3:1 ratio in F_2. The test cross progeny of the F_1 heterozygotes yielded the expected 1:1 ratio, confirming monofactorial recessive pattern of inheritance, thus indicating that these mutants are recessive and autosomal.

The mutations mentioned here and others reported by us elsewhere (Chowdaiah and Shetty, 1976; Shetty and Chowdaiah, 1976)[3,4] which are a part of this continuing search for a better understanding of the genetic system of *Culex fatigans* offer interesting possibilities for future work.

This work was supported in part by a grant from the Indian Council of Medical Research.

Department of Zoology,
Bangalore University,
Bangalore 560001, *July* 31, 1976.

N. JAYA SHETTY.
B. N. CHOWDAIAH.

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1. Laven, H., In: *Genetics of Insect Vectors of Disease* (Eds. J. W. Wright and R. Pal), Elsevier Pub. Co., 1967, p. 17.
2. Shetty, N. J., *Ph.D. Thesis*, Bangalore University, 1974.
3. Chowdaiah, B. N. and Shetty, N. J., *Jour. Genet.*, 1967, 62, 131.
4. Shetty, N. J. and Chowdaiah, B. N., *Jour. Heredity* (in press).

Discovery of Fossiliferous Eocene Rocks from Shankarpur, Tehri District, Uttar Pradesh

The Subathu beds Eocene are the only known marine sediments of Teritiary age in Himalaya. They have been found to occur in two structural settings, namely, (i) Autochthonous Eocene belt occurring to the north of the Siwalik sediments and separated from it by the well-known tectonic feature known as Main Boundary Fault. It attains best development towards west of Subathu-Bilaspur area of Himachal Pradesh and thins out towards south-east and is considered to disappear about 20 kms. east of Nahan (Pascoe, 1962), (ii) isolated patches of Eocenes occurring over the rocks of Nappe zone and also as windows within this zone. Between Dehra Dun and Rishikesh, it has been reported to occur in two tectonic windows, Bidhalna and Pharat (Auden, 1936).

Recent work in Rishikesh area has revealed the presence of a belt of rocks comprising dark grey carbonaceous shales, quartzites, grey silliceous limestone, greenish and purplish shales, colitic and sandy limestone. The colitic limestones are fossiliferous and contain broken shellfragments of pelecypod and corals, etc.

This belt is exposed between Shankarpur (30° 10′ 10″ : 78° 15′ 05″) in the west to Bidasni (30° 59′ : 78° 18′) in the east. Further eastward extension of this belt is yet to be traced. The fossiliferous limestone occurs in a number of bands interbedded with purple and green shales. To the south, this belt is bounded by the Main Foundary Fault, south of which Siwaliks are exposed. In the north, it is overlain by the rocks of Krol Nappe.

Presence of fossiliferous limestone in this belt together with analogous structural disposition with Subathus of the Main belt in Himachal Pradesh, suggests that it may be the continuation of the autochthonous Eocene belt. Detailed work is in progress. In view of the importance of Tertiary sediments for oil exploration in the foot-hill zone of Himalaya, the discovery of these rocks is thus of considerable significance.

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Geological Survey of India, G. KUMRAR.
Northern Region, J. N. DHAUNDIYAL.
11-B, H Road, Mahanagar Extension,
Lucknow 226006, *August* 16, 1976.

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1. Auden, J. B., *Rec. Geol. Sur. Ind.*, 1936, p. 71.
2. Pascoe, E., *A Manual of Geology of India and Burma*, Pt. III, 1962.

A New Disease of 'Kopak' (*Celba pentandra* Gaertn.) Caused by *Colletotrichum gloeosporioides* (Per. ex Fr.) von Arx

During the month of September and October 1975–76 many of the plants grown in nursery garden of Horticulture, Agricultural University, Jabalpur, showed severe leaf-spot and die-back symptoms. The disease affected the plants at all stages of growth but is very destructive in young stages. The size of the spots increased with age and finally two or more spots coalesced developing big necrotic areas covering nearly the entire leaf-let. Leaf scars were generally found to be the site of infection. Isolations from diseased portions of stems, leaves and petioles always yielded a species of *Colletotrichum*.

Pathogenicity tests were carried out on three to four-month old plants by spraying an aqueous suspension of conidia from 15 days old culture for 48 hours at high humidity. Suitable controls were also maintained. The disease symptoms appeared within 8 days. A similar *Colletotrichum* species was recovered from the artificially produced disease.

The characters of conidia of the *Ceiba* isolate are in agreement with those of *C. gloeosporioides* (Pers. ex Fr.) von Arx[1]. The causal organism is identified *C. gloeosporioides* (Pers. ex Fr.) von Arx[1] and Rajak[2].

This is the first report of this fungus causing disease of 'Kopak'. The culture has been deposited in Plant Pathology Department, J.N. Agricultural University, Jabalpur.

Department of Plant Pathology, N. D. SHARMA.
J. N. Agricultural University, A. C. JAIN.
Jabalpur–482 004, M.P., India.
June 23, 1976.

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1. von Arx, J. A., *Phytopath. Z.*, 1957, **99**, 415.
2. Rajak, R. C. "Studies on the morphology and physiology of some Indian species of the genus *Colletotrichum*," *Ph.D. Thesis*, Jabalpur University, Jabalpur, 1972.

Occurrence of *Phoma exigua* Desm. on Sunflower

During *Kharif* 1975 numerous dark-brown to black lesions of various sizes and shapes were found on older leaves of sunflower in association with those caused by *Alternaria helianthi*. The surface of these lesions was rough and had a dark-grey or black sheen. Isolations from such lesions consistently yielded a pycnidial fungus which was identified as *Phoma exigua* Desm. (C.M.I. Acc. No. I.M.I. 1986 21). This isolate does not produce any pigment or crystal on Malt-agar plates.[1]

Phoma oleracea var. *helianthi-tuberosi*[3] and another species of *Phoma*[2] are known to attack sunflower. Pathogenicity test conducted with the isolate on 1-month old seedlings of sunflower (EC 68413) at 25°-30°C under humid conditions showed that the fungus is a weak parasite, capable of producing only minute lesions on leaves but readily enters the host through bruishes or wounds. This is the first report of *Phoma exigua* occurring on sunflower from India. A culture of this fungus has been deposited with the Indian Type Culture Collection, I.A R I., New Delhi, under Acc. No. 2062.

We are grateful to Dr. A. Johnston and Dr. E. Punithalingam, C.M.I.. Kew, England, for identification of the fungus. Thanks are also due to Dr. J. N. Kapoor and Prof. V. V. Chenulu, for useful criticism and keen interest in the work.

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Indian Agricultural Res. Inst.,
New Delhi 110012,
India, *July* 10th, 1976.

T. P. Bhowmik.
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1. Boerema, G. H. and Howeler, L. H., "*Phoma exigua* Desm. and its varieties," *Persoonia*, 1967, **5**, 15.
2. Frezzi, M. J., "A species of the genus *Phoma* as parasite of sunflower in Manfredi," *Publnes Estac. Exp. agric. Manfredi*, 1964, **19**, 37.
3. McDonald, W. C.,‘ ‘*Phoma* black stem of sunflower," *Phytopathology*, 1964, **54** 492.

Two Additional Hosts of *Alternaria helianthi*

Alternaria helianthi is known to cause a severe leaf spot disease of Sunflower (*Helianthus annuus* L,) resulting in a appreciable loss in yield, seed weight etc.[1,2]. In a study on reaction of large number of sunflower germ plasm materaial and other related species, it was observed that *H. debelis* and *H. argophyllus* to be infected by *A. helianthi*. *H. debelis* was highly susceptible throughout its growth period while *H. argophyllus* was highly resistant and infection occurred only under prolonged warm humid conditions. A normal feature observed in the case of *H. argophyllus* was, that, the foci of infection was generally leaf axil, resulting in the death and drooping of the leaf, although, the leaf spot symptom was not uncommon. The two isolates of *A. helianthi* from the above hosts did not differ in any way from the sunflower isolate and were cross inoculable.

Thanks are due to Dr. H. C. Govindu, Senior Professor, for facilities.

AICRP on Sunflower
University of Agricultural Sciences,
Hebbal, Bangalore-560024,
September 20, 1976.

T. B. Anilkumar.
M. N. L. Sastry.
A. Seetharam.

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1. Anilkumar, T. B., *et.al.*, *Curr.*, *Sci.*, 1974, **43**, 93.
2. Anonymous, *Annual Report*, AICORP (Sunflower), U.A.S., Bangalore, 1976,

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REVIEWS AND NOTICES OF BOOKS

Handbook of Mycology, Part II : Basidiomycetes and Fungi-Imperfecti. (2nd edition). By M. N. Kamat. (Prakash Publishing House, Pune 411 002), 1976. Pp. 1–165, 25 × 19 cm. Price Rs. 15.

After a frontispiece showing the photographs of eminent mycologists and plant pathologists and a preface to the first and second editions, a general account of the Basidiomycetes is followed, which includes laboratory exercises; next an account of the Fungi-Imperfecti on the same model, like topics and laboratory exercises are given. In the appendices, there is an account of Lichens, Keys to Important Genera, Monographs, Reviews and related topics. As the author states, this second edition has been primarily enlarged to meet the needs of Indian students and teachers and has been comprehensively revised and enlarged to include new figures and plates. In general, the treatment is satisfactory and useful to the students. The line drawings form an additional feature of this book.

Since the bias given is for a text-book, and though the author expresses regrets for 'printer's devil', this cannot be accepted. It is seen that there are a number of spelling mistakes and adequate care should have been taken for careful editing and proof-reading. Even the initial of such a well-known mycologist like Dr. M. J. Tirumalachar is wrongly given. Further, such references as Boss (1927), Brown (1940), Kulkarni (1965), Rajendran (1969) are not cited in the reference. The citation of literature is very unsatisfactory and the abbreviations of journals should have been strictly according to the World List of Periodicals. For reference No. 73 on page 28, no particulars are given and for reference No. 79 on the same page, the year of publication is not given. Again, in the keys to important genera, no uniformity has been maintained and the keys would have been appropriate and meaningful if the dichotomous method was strictly followed. A list of Errata if included would have considerably brought down these shortcomings to some extent.

K. SUBRAMANYAM.

Marine Science Communications (Vol. I, No. 1). (Marcel Dekker, Inc., 270, Madison Avenue, New York 10016, N.Y.), 1975. Pp. 100, Annual Subscription : $ 35.00 per volume, single issue $ 7.50, Postage and handling charges for outside U.S. $ 6.50.

Environmental protection has become a key concept at present and the major focus of marine policy for the seventies is the concern for the preservation and improvement of the marine environment. The need for extensive knowledge of marine environment is now very much greater than ever before and is very pressing. The marine sciences have considerable practical justification and also afford an excellent introduction to scientific method and to interdisciplinary scientific enterprise. Necessarily therefore marine science should have a loose partnership of disciplines and the results from such an interdisciplinary approach naturally complement and catalyse each other, thus establishing a healthy tradition of bringing out interdisciplinary information.

A welcome addition, therefore, is *Marine Science Communications,* which is devoted to basic, applied and ocean systems research, presenting a broad spectrum of information to marine scientists. Six scientific articles published in Vol. I, No. 1 represent very well the above objective and are of high quality.

A very gratifying fact about the journal is publication of articles within 3 to 4 months' time from the date of receipt if accepted without modification. This is an excellent incentive and the International Educational Board deserves our full support.

There is no doubt that this scientific journal, of excellent quality and production, will be found to be extremely useful by all marine scientists.

R. NATARAJAN.

6126/76. Published by S. R. S. Sastry, Manager, Current Science Association, Bangalore-560006
and Printed at The Bangalore Press, Bangalore-560018.

DOUBLE STRANDED POLYNUCLEOTIDES: TWO TYPICAL ALTERNATIVE CONFORMATIONS FOR NUCLEIC ACIDS

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ABSTRACT

Two typical alternative conformations for double stranded polynucleotides with Watson-Crick base pairing scheme are presented. These types avoid tangling of the two chains. Representative models of these types with two different views, to show the similarity and dissimilarity between these models and the Watson-Crick model, are given.

INTRODUCTION

THE well-known Watson and Crick model[1] for DNA is a right handed double helix. Our extensive work, carried out on the analysis of helical conformations for double stranded polynucleotides with antiparallel chains revealed that both right and left handed double helices are stereochemically possible. Based on our analysis, a type I model, employing Watson-Crick base pairing and conformations for the nucleotides, as observed in single crystals, was presented* in which the polynucleotide chain had both right and left handed segments. This immediately avoids the tangling of the two chains and hence has, in addition, other advantageous features over the Watson-Crick model. Since then a detailed conformational analysis carried out by using an IBM 360/44 computer has revealed two typical conformations based on the orientations of the sugars of the chain. These two types of conformations are energetically more favourable than the Watson-Crick double helix. The present paper outlines these two alternative conformations.

RIGHT AND LEFT HANDED DOUBLE HELICES

Figure 1 (*a*) shows the schematic diagram for right and left handed double helices (with antiparallel chains) with base pairing shown as dotted lines. The dihedral angles (which are restricted)[2] about the various single bonds in the backbone and the side chain (base) dihedral angle about the glycosidic bond are shown in Fig. 1 (*b*). The regions of backbone dihedral angles for the possible right and left handed double helical conformation have already

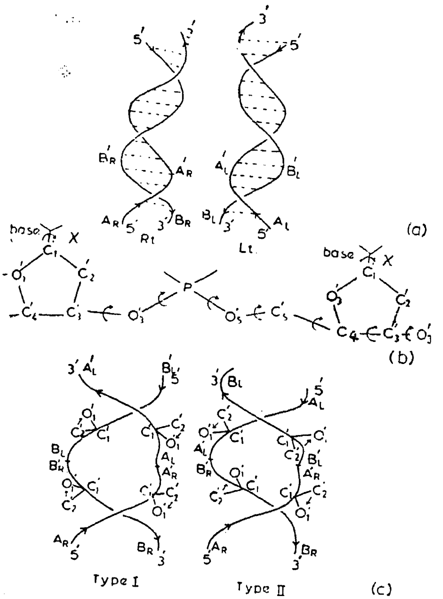

FIG. 1. (*a*) Schematic diagram of right and left handed helices; (*b*) Polynucleotide backbone; (*c*) The two types of linking right and left segments

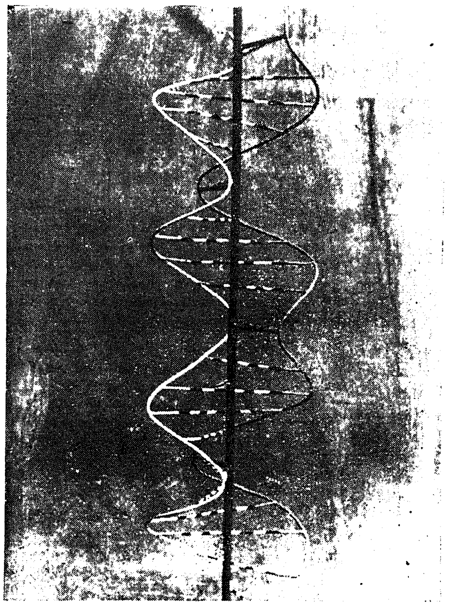

FIG. 2. Shematic view of the models (Types I and II) to show lateral separation of the two chains.

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* At the Divisional Review of the Chemical and Biological Sciences, Indian Institute of Science, Bangalore, February 23, 1976.

been given[2]. Further analysis of the regular helices with antiparallel chains and with the number of residues per turn $n \simeq 10$ and height per turn $h \simeq 3 \cdot 4$, restricted these possibilities to a few. Suffice it to say, that sterically both left and right handed conformations can be obtained with *gg*, *gt* and *tg* conformations about the C_4'–C_5' bond with the corresponding conformations about the P–O bonds. Most interestingly, our analysis of the conformation about the glycosidic bond (for a nearly perpendicular orientation of the bases as in B DNA) indicates two alternating regions for right and left handed helices. These are (*a*) for the right handed helix (1) normal *anti* region $\chi < 90°$ and (2) *syn* region $180° < \chi < 240°$, and (*b*) for the left handed helix (1) low *anti* region $\chi \lesssim 0°$ and (2) high *anti* region $\chi > 90°$. The exact value in each case would depend on the backbone dihedral angles as well as the puckering of the sugar. The flexibility of the sugar puckering was taken into account as described earlier[2]. It may be worthwhile to mention here that right and left handed helices can have the same radius for the phosphorus atom ($\sim$ 9 Å) but at the same time have different radii for the C_1' atom. In such a case the radius of C_1' atoms in the left handed helix is larger by 0·5 Å than the radius of the C_1' atoms in the right handed helix. However, the Fourier transforms of both the models are very similar. The other features of the right and left handed double helices will be discussed in greater detail elsewhere.

THE TWO TYPES OF CONFORMATIONS : I AND II

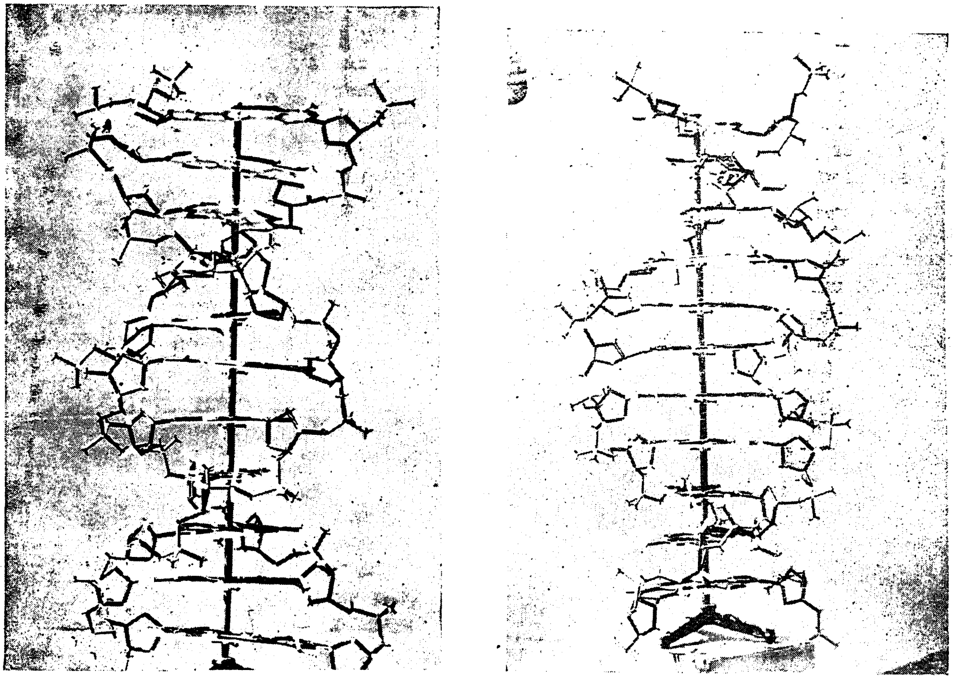
There are two typical ways [Fig. 1 (*c*)] by which the right and left handed segments can be joined to form a double stranded polynucleotide chain having Watson-Crick base pairing but which would avoid tangling. For a repeating unit of ten base pairs, roughly half a turn of the right handed double helix can be linked to the corresponding half a turn of the left handed double helix. In type I, the right handed segments A_RA_R' and B_RB_R' are joined to the left handed segments A_LA_L' and B_LR_L' respectively. If the left and right handed segments are joined this way, the orientations of the sugars of the chain are in the same direction (shown in the diagram by the vectors $C_2' \rightarrow O_1'$). In the type II structure, the right handed segments A_RA_R' and B_RB_R' are joined to the left handed segments $B_L'B_L$ and $A_L'A_L$ by an inversion of the left handed segment such that the chain directions 5′ to 3′ and 3′ to 5′ are preserved. In such a structure the sugars in the right handed segment are pointing roughly opposite to the sugars in the left handed segment (shown by the vectors $C_2' \rightarrow O_1'$).

Both the type I and type II structures involve 'folds' or 'bends' along the chain and these folds occur in pairs approximately after every five base pairs. Since there is flexibility about the backbone of the polynucleotide chains[2], there can be a family of folds. In the representative models for each type shown, we have used the types of fold which we consider as plausible. Apart from the types of folds, the main feature of both the structures is the value of χ for the base pair. In the type I structure, if a normal *anti* conformation ($\chi < 90°$) is used for the right handed segment, then the left handed segment has a value for $\chi > 90°$ (high *anti* region). In type II, for the normal *anti* conformation of the right segment, the left segment has $\chi \lesssim 0$ (low *anti* region). Similarly, for the *syn* conformation of the bases in the right handed segment, both types I and II are possible, but with the χ values interchanged for the left handed segments. The *syn* conformation was not investigated further.

Thus in both types I and II, the two polynucleotide chains have a wavy structure and avoid intertwining (Fig. 2). Both have alternating right and left segments along the length of the chain and have a pair of folds in each repeating unit. In type I, the sugars point roughly in the same direction in both the segments which have normal bases stacked. In type II, the sugars in the adjoining segments are inverted ; that is, point in opposite directions. This structure has the base in the adjoining segments in inverted positions and stacked accordingly.

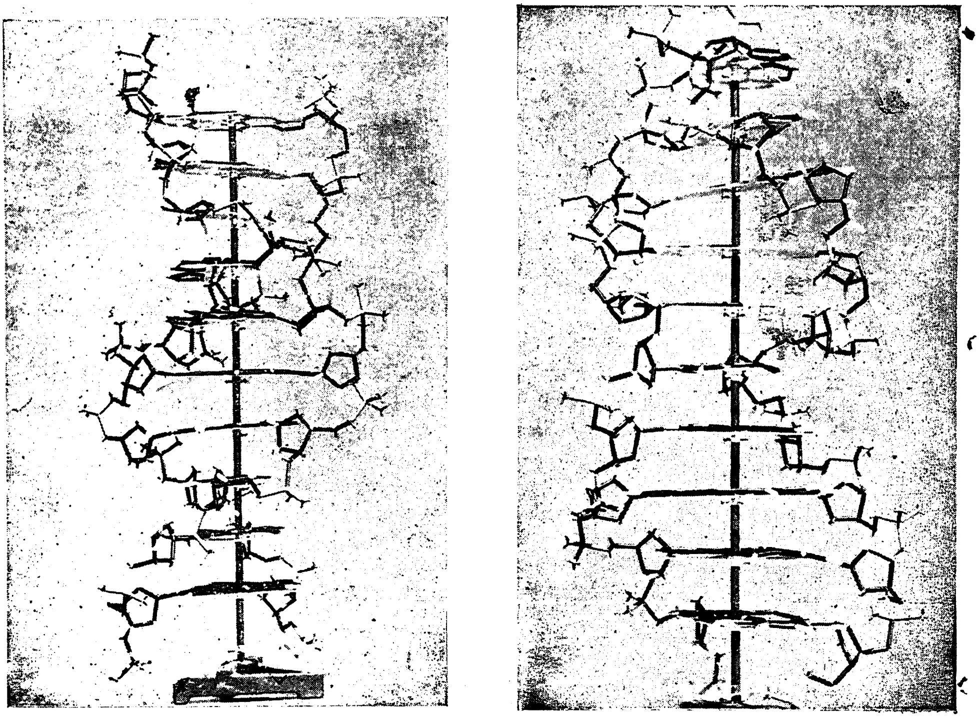
THE NEED FOR FOLDING

It is clear that as the conformational energy difference between the various types of backbone conformations of the monomer unit of the polynucleotide chain is small[3,4], the conformations of the chains are mainly determined by the base-base interactions apart from hydrogen bonds. As the Watson-Crick base pairing has been maintained in our models, the essential difference between our two structures and the Watson-Crick model lies in the mode of stacking of bases. The stacking energy between any two bases in different orientations and when separated by various distances was therefore investigated. A detailed report is under preparation (Sasisekharan and Gautam Gupta). The results of interest to us are the following : In the case of cytosine (C), stacking is achieved only for interactions between normal C and inverted C. The normal base-base interaction is repulsive and becomes attractive only when one C inverted. This agrees with the calculations reported earlier[5] ; similar conclusions hold for G–G interactions. Thus a G–C pair followed by another G–C pair will be highly stable when one of the G–C pairs is inverted ; the energy difference is more than 10 kcals/mole. Thus a C–C (or G–G) base sequence along the polynucleotide chain can bring about a fold in type II.



(3 *a*) (3 *b*)

FIG. 3. A Model of type I—Two views.



(4 *a*) (4 *b*)

FIG. 4. A Model of type II—Two views.

For A–A, both (normal base)–(normal base) interactions and (normal base)–(inverted base) interactions have nearly the same energy. For T–T, the normal (base)–(base) interaction is small; however, it is lower than the interaction energy between a normal base and an inverted base.

In the case of purine-pyrimidine (or pyrimidine-purine) sequences (normal base)-(normal base) stacking is favoured. However, depending upon the sequence, either a right or a left stacking of bases is preferred. Thus, in a sequence GpC, the stacking is right handed whereas in CpG the stacking is left handed. As a consequence, sequences like CpG can involve a right to left fold in type I. Similarly, for ApT and TpA respectively, right and left stacking of bases can occur; however, the energy difference is small.

REPRESENTATIVE MODELS OF TYPES I AND II

A representative model for type I with two different views is shown in Fig. 3. Here, we have used the *gg* conformation about the C_4'–C_5' bond for the right handed segment of the chain[6] and *gt* conformation for the left handed segment of the chain. The *gt* conformation has already been observed in a crystal structure[7]. It may be worthwhile to mention that the *gg* conformation for the left handed segment is also possible for this model. As regards the two types of folds, one involves a change from *gg* to *gt* conformation about the C_4'–C_5' bond as has been used in the kinky helix[8]. The other involves a change from *gg* to *tg* conformation. Again, the *tg* conformation has also been observed in a crystal structure[9]. For the right handed segment, the C_2' endo conformation for the sugar has been used. For the left handed segment the C_3' endo sugar has been used.

A model for type II with two different views is shown in Fig. 4. The conformation employed about the C_4'–C_5' is *gg* for both the right and the left handed segments. The sugar puckering employed is C_2' endo in the right handed segment and C_2' endo –C_1' exo for the left segment. Here one fold is achieved by employing g^+g^+ conformation about the P–O bonds. This conformation has been observed in two crystal structures[10,11]. The other fold is achieved by slight modifications of the conformations about the various single bonds that are employed for the right handed segment. One view of both the types resembles the typical Watson-Crick model. However, the other view, taken at 90° from the first one, reveals marked differences and clearly shows the lateral separation of the two chains without intertwining.

Other types of puckering could be employed in both these models. Also, the puckering of the sugars at the folds could be different. In both the models we have maintained the radius of the phosphorus atom to be very nearly 9 Å. This could also be varied if necessary, and the width of the two segments could also differ.

The Fourier transform of these models has not been computed, as the mathematics of the same has to be worked out. However, the Fourier transforms of the corresponding left handed and right handed helices are similar. It is clear that these types of structures could be built with folds at other levels and not necessarily after every five base pairs. These types of arrangements have more degrees of freedom and hence flexible. As the models presented here are tentative, we do not wish to discuss their geometrical properties. Full details of the two structures will be published elsewhere.

CONCLUSIONS

The purpose of the present paper is to show that double-stranded polynucleotide chains having base orientation as in B DNA can be constructed in two typical ways without tangling. The two types of arrangements appear to bend more readily than the conventional double helix. The preliminary model building indicates that a minimum of three base pairs are necessary between any two folds. In a nucleic acid chain, where we have a sequence of bases, we believe that both the types could be present in a single molecule. In such a case DNA need not be a double helix throughout and such a structure has a number of attractive features and these will be dealt with elsewhere.

Note: A paper "A possible conformation for double-stranded polynucleotides" has just appeared in the *Proceedings of the National Academy of Sciences*, USA, Vol. 73, No. 9, 1976, pp. 2959-2963) which describes a model similar to the type I structure presented here. We thank Professors H. Sharat Chandra and O. Siddiqi for drawing our attention to this report.

ACKNOWLEDGEMENTS

We wish to thank Prof. G. N. Ramachandran for his keen interest. We thank the Science and Engineering Research Council, of the Department of Science and Technology, Government of India, for financial support. One of us (N. P.) acknowledges financial assistance from the University Grants Commission in the form of a Junior Research Fellowship.

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1. Watson, J. D. and Crick, F. H. C., *Nature*, 1953, 171, 737.
2. Sasisekharan, V., *The Jerusalem Symposium on Quantum Chemistry and Biochemistry*, Vol. V, 1972, p. 247.
3. Lakshminarayanan, A. V. and Sasisekharan, V., *Biopolymers*, 1969, 8, 489.

4. Sasisekharan, V. and Lakshminarayanan, A. V., *Ibid.*, 1969, 8, 505.
5. Pullman, A. and Pullman, B., *Advances in Quantum Chemistry*, 1968, 4, 267.
6. Arnott, S., Dover, S. D. and Wonacott, A. J., *Acta Cryst.*, 1969, B 25, 2192.
7. Seshadri, T. P. and Viswamitra, M. A., *Pramāna*, 1974, 3, 218.
8. Crick, F. H. C. and Klug, A., *Nature*, 1975, 255, 530.
9. Viswamitra, M. A., Seshadri, T. P. and Post, M. L., *Ibid.*, 1975, 258, 542.
10. Suck, D., Manor, P. C., Germain, G., Schwalbe, C. H., Weimann, G. and Saenger, W., *Nature New Biology*, 1973, 246, 161.
11. Rubin, J., Bernnan, T. and Sundaralingam, M., *Biochemistry*, 1972, 11, 3112.

KINETICS OF DECOMPOSITION OF NITROUS OXIDE OVER NICKEL TITANATE

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ON account of their greater chemical stability, structural reproducibility and catalytic activity than their parent binary oxides, ternary oxides of transition metals have generated wide-spread interest in recent years as catalysts of choice for a diverse variety of catalytic reactions. As part of a general study of the catalytic properties of these oxides, the kinetics of decomposition of nitrous oxide were studied on a series of titanates, with a view to evaluating the usefulness of this reaction for comparative assessment of their catalytic activities for oxidation reactions. No report is available of any previous study of this reaction over ternary oxides. The results obtained with nickel titanate are reported in the present communication. Similar results were obtained with the titanates of cobalt, manganese, magnesium, barium, strontium and calcium.

Nickel titanate was prepared by the method of Saikali *et al.*[1]. Its composition ($NiTiO_3$) and ilmenite structure were established by chemical analysis and X-ray diffraction. BET surface area determined with nitrogen at $-198°$ C $= 3 \cdot 14$ m^2/gm. Nearly pure N_2O, was further purified by selective freezing at $-78°$ C.

The kinetics of N_2O decomposition over nickel titanate were studied in the temperature range 440–510° C in a quartz reactor, with closed circuit recirculation using an all-glass electromagnetic pump. The decomposition rate was followed by noting the increase in pressure as a function of time. After each kinetic run, the catalyst was "prepared" for the next run, by evacuation at 520° C for 6 hours followed by 8 hours exposure to oxygen (100 torr) at the temperature of the next run and a short evacuation at the same temperature for 3 minutes. This procedure for surface restoration was found to yield reproducible results in repeat runs.

Winter[2], who studied the decomposition of nitrous oxide over rare earth oxides, noted that different rate equations were to be used to express the kinetics of the decomposition at high and low pressures, namely,

$$-\frac{dP_{N_2O}}{dt} = \frac{kP_{N_2O}}{(P_{O_2})^{1/2}}, \quad \text{at } N_2O \text{ pressures} > 200 \text{ torr} \qquad \text{(I)}$$

and

$$-\frac{dPN_2O}{dt} = k'.P_{N_2O}, \quad \text{at } N_2O \text{ pressures} < 50 \text{ torr} \qquad \text{(II)}$$

The same duality of kinetic behaviour at high and low N_2O pressures was observed in the present work also, as may be seen from the consistency of the k values (low standard deviations) calculated with the appropriate rate expressions (*vide* Table I).

TABLE I

Values of rate constants deduced from differential equations I and II

Temp. °C	Values of rate constants (k)			
	k ($cm^{1/2}$ min^{-1} m^{-2}) $\times 10^{+4}$ (above 200 Torr)		k (min^{-1} m^{-2}) $\times 10^{+4}$ (below 50 Torr)	
	Equation I	Equation II	Equation I	Equation II
440	$2 \cdot 24 \pm 0 \cdot 182$	$5 \cdot 44 \pm 1 \cdot 71$	$3 \cdot 66 \pm 0 \cdot 76$	$9 \cdot 7 \pm 0 \cdot 036$
460	$3 \cdot 85 \pm 0 \cdot 1$	$6 \cdot 59 \pm 1 \cdot 41$	$4 \cdot 37 \pm 0 \cdot 92$	$13 \cdot 7 \pm 0 \cdot 7$
490	$5 \cdot 29 \pm 0 \cdot 1$	$4 \cdot 49 \pm 0 \cdot 8$	$7 \cdot 76 \pm 1 \cdot 85$	$15 \cdot 7 \pm 0 \cdot 04$
510	$8 \cdot 3 \pm 10 \cdot 27$	$11 \cdot 9 \pm 2 \cdot 92$	$12 \cdot 99 \pm 4 \cdot 14$	$27 \cdot 1 \pm 0 \cdot 11$

TABLE II

Comparison of rate constants deduced from differential and integral rate equations

Temp. °C	Values of rate constant (k)			
	Above 200 Torr		Below 50 Torr	
	From Eqn. I $\times 10^{+4}$	From Fig. 1 $\times 10^{+4}$	From Eqn. II $\times 10^{+4}$	From Fig. 2 $\times 10^{+4}$
440	2·24	2·2	9·7	10·1
460	3·85	3·82	13·7	14·0
490	5·29	5·57	15·7	15·7
150	8·31	8·27	27·1	27·5

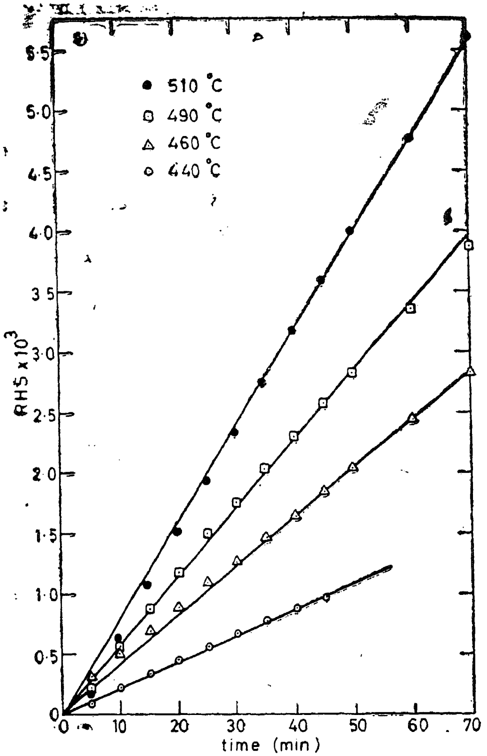

FIG. 1. Plots of Equation I′ for decomposition at pressures > 200 Torr.

This could also be demonstrated by using the integrated forms of the two rate-expressions[3], namely,

$$kt = (P_1 + P_2)^{1/2}\left\{\ln\left[\frac{(P_1 + P_2)^{1/2} + (P_1 + P)^{1/2}}{(P_1 + P_2)^{1/2} - (P_1 + P)^{1/2}}\right] - 2\frac{(P_1 + P)^{1/2}}{(P_1 + P_2)^{1/2}}\right\}, \qquad (I')$$

where $2P_2$ = initial pressure of N_2O (> 200 torr), $2P$ = pressure of N_2O decomposed at time t, P_1 = initial pressure of O_2, and

$$\log P_{N_2O}\ (< 50\ \text{torr}) = -\frac{k'\,t}{2\cdot303} + C. \qquad (II')$$

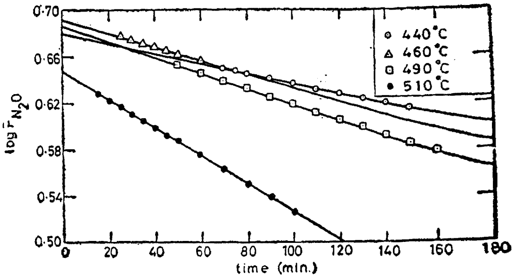

FIG. 2. Plots of Equation II′ for decomposition at pressures < 50 Torr.

Plots of the experimental data substituted in these equations are shown in Figs. 1 and 2, respectively. The linearity of these plots and the closer agreement between the value of the rate constants (k) deduced from the two sets of equations, shown in Table II, confirm the validity of these equations in the respective pressure regimes.

TABLE III

Kinetic parameters

Catalyst	E_a (kcals/mole)	k_0 ($cm^{1/2}$ min^{-1} m^{-2})	k at 500 °C ($cm^{1/2}$ min^{-1} m^{-2})
$NiTiO_3$	18·14	24·4	$8\cdot65 \times 10^{-4}$
NiO	39·5	198·3	$7\cdot46 \times 10^{-7}$
TiO_2	39·0	0·928	$1\cdot16 \times 10^{-11}$

The values of the apparent activation energy (E_a) and the frequency factor (k_0) for N_2O decomposition on $NiTiO_3$, NiO and TiO_2, above 200 torr in the temperature range 300–360° C for NiO and 440–510° C for $NiTiO_3$ and TiO_2 and the k values at 500° C (computed temperature) are given in Table III. The higher catalytic activity of $NiTiO_3$ relative to its constituent binary oxides, *i.e.*, the operation of a catalytic synergetic effect, is evident from these data.

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1. Saikali, Y. and Paris, J. M., *C.R. Acad. Sci.*, Paris, Serc., 1967, **265** (19), 1041.
2. Winter, E. R. S., *J. Catalysis*, 1969, **15**, **144**.
3. —, *Discussions Faraday Soc.*, 1959, **28**, **183**.

SYSTEMATICS IN TOTAL (*n*, 2*n*) CROSS SECTIONS AT 14 MeV

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ABSTRACT

The 14–15 MeV (*n*, 2*n*) cross sections are found to depend mainly on the asymmetry parameter and the Q-value. No shell effects are found to exist in these cross sections. The total (*n*, 2*n*) cross sections are found to be well predicted by an empirical relation which takes into account the Q-value and the asymmetry parameter in addition to the geometrieal cross section.

THERE have been some attempts[1-4] on the systematic study of the (*n*, 2*n*) cross sections at 14–15 MeV energy of the incident neutrons. Bormann[1], Strohal *et al.*[4] and Chatterjee *et al.*[3] describe the shell effects to be predominant effect in (*n*, 2*n*) cross sections, whereas Hille[2] has observed no such effects. He has found a strong dependence of the (*n*, 2*n*) cross sections on the asymmetry parameter. We have, however, observed the dependence of the (*n*, 2*n*) cross sections on both the Q-value of the (*n*, 2*n*) reaction and the asymmetry parameter.

We have taken into consideration all the available experimental data[5-8] of the (*n*, 2*n*) cross sections. In order to see the neutron and proton shell effects clearly, target nuclei with Z or N equal to 2, 6, 8, 14, 20, 28, 40, 50, 64, 82, 100 and 126 have been chosen for which there exist the total (*n*, 2*n*) cross sections in literature. The (*n*, 2*n*) cross sections of these nuclei and the corresponding asymmetry parameter have been plotted *vs.* mass number A of the target nucleus on the same plot as shown in Fig. 1. The vertical bars in this plot include the reaction cross section values reported by different authors for each case. It is obvious from this plot that with the increase in A, it is the asymmetry parameter which influences the (*n*, 2*n*) cross section. The simultaneous effect of the Q-value and the asymmetry parameter is manifested in plots (*a*), (*b*) and (*c*) of Fig. 2, where the (*n*, 2*n*) cross sections are plotted *vs.* atomic number Z of the target nucleus for three neutron numbers of 28, 50 and 82. All these cases show that as the Q-value becomes more and more negative and also as the asymmetry parameter decreases, the (*n*, 2*n*) cross section decreases accordingly. No shell

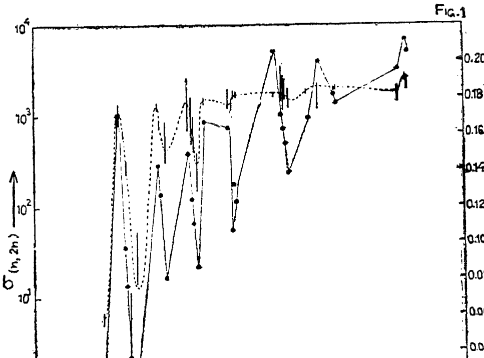

FIG. 1. Experimental total (*n*, 2*n*) cross sections at 14–15 MeV (solid line) represented by vertical bars and the corresponding asymmetry parameter (dotted line) *versus* mass number A of the target nucleus with Z or N equal to 2, 6, 8, 14, 20, 28, 40, 50, 64, 82, 100 and 126. The curves are arbitrary lines showing the trend.

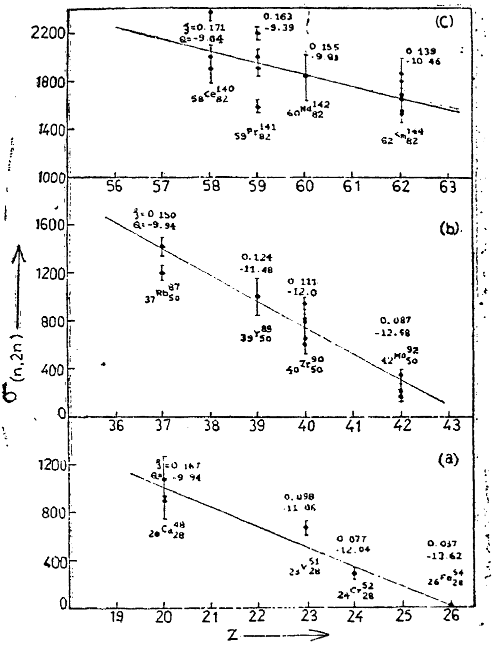

FIG. 2. Experimental total (*n*, 2*n*) cross sections at 14–15 MeV *versus* the proton number Z of the target nucleus. Plots (*a*), (*b*) and (*c*) correspond to the neutron number 28, 50 and 82 respectively. Q-value and asymmetry parameter of each target nucleus is given in the figure.

effects are observed corresponding to the magic proton numbers. Clearly the Q-value and the asymmetry parameter affect the $(n, 2n)$ cross sections.

The strong dependence of the $(n, 2n)$ cross sections on the asymmetry parameter and the Q-value aroused the need to formulate the following empirical relation for the total cross section value at 14–15 MeV :

$$\sigma_{(n, 2n)} = \pi R^{20} (A^{1/3} + \bar{\lambda})^2 \quad [\exp Q \{0 \cdot 25\, \xi - 0 \cdot 36\, \xi^2 - \gamma\}] \qquad (1)$$

For 14–15 MeV neutrons $\bar{\lambda} = 1$ and with $R_0 = 1 \cdot 2f$, the geometrical cross section $\pi R_0{}^2 (A^{1/3} + \bar{\lambda}^2)$ becomes $45 \cdot 2\ (A^{1/3} + 1)^2$ and so relation (1) can be re-written as :

$$\sigma_{(n, 2n)} = 45 \cdot 2\, (A^{1/3} + 1)^2 \quad [\exp Q \{0\ 25\, \xi - 0 \cdot 36\, \xi^2 - \gamma\}] \qquad (2)$$

where Q = Q-value of the $(n, 2n)$ reaction

ξ = Asymmetry parameter

and γ is a constant which is different for different ranges of the asymmetry parameter as listed below :

$\gamma = 0 \cdot 36$ for $0 \leqslant \xi < 0 \cdot 040$

$\gamma = 0 \cdot 24$ for $0 \cdot 040 \leqslant \xi < 0 \cdot 063$

$\gamma = 0 \cdot 10$ for $0 \cdot 063 \leqslant \xi < 0 \cdot 087$

and

$\gamma = 0 \cdot 033$ for $\xi \geqslant 0 \cdot 087$

This empirical relation predicts the total $(n, 2n)$ cross sections within 10% for all nuclei with a very few exceptions in the case of low ξ-values.

1. Bormann, M., *Nucl. Phys.*, 1965, 65 **A**, 257.
2. Hille, P., *Ibid.*, 1968, 107 **A**, 49.
3. Chatterjee, S. and Chatterjee, A., *Ibid*, 1969, 125 **A**, 593.
4. Strohal, P., Cindro, N. and Eman, B., *Ibid.*, 1962, 30, 49.
5. Kondaiah, E., *T.I.F.R. Preprint No. NP*, 1974, 1.
6. Kao, T. H. and Alford, W. L. *Nucl. Phys.*, 1975, 237 **A**, 11.
7. Qaim, S. M., *Ibid.*, 1974, 224 **A**, 319.
8. — and Grace, C., *Ibid.*, 1975, 242 **A**, 317.

SHORT PERIOD WAVES IN THE F2-REGION DURING MAGNETICALLY DISTURBED PERIOD

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THE acoustic waves in the F-region occur either due to natural sources[1-3] or due to artificial reasons[4,5]. The aim of this report is to present an evidence for the presence of short period waves around '1' minute in the F2-region of the ionosphere over Waltair. These waves are found to occur on magnetically disturbed days. This is the first time that such an observation is made.

The investigations are carried out by taking phase path measurements of F2-region echoes at vertical incidence on 5·6 MHz pulsed signal at Waltair (Geographic 17° 43′ N, 83° 18′ E, Geomag. 7° 24′ N). The experimental details and the method of analysis of the data are similar to those adopted by Reddy and Rao[6,7]. The positive slope of the fringes in the records illustrated in Fig. 1 indicates an increase in phase path and *vice versa*. The fringe pattern in the bottom part of the record corresponds to *'x'* magnetoionic component and the upper part relates to that of *'o'* as indicated. The local times are indicated.

The samples of phase path records shown in Fig. 1 from (*a*) to (*g*) are reproduced from the record of F2-region echo, obtained on 19–4–1974. Originally this record commenced at 1440 hrs. and the echo had both *'o'* and *'x'* components slightly resolved at a virtual height of about 315 km. Phase path variations between 1440–1520 hrs. are smooth and continuous with a few quasi-periodic oscillations of larger periods (due to regularly observed TIDs) superposed on a linearly increasing phase path. However during this interval, the sequence of short period oscillations is also indicated by just perturbing the smooth variattions as shown in sample (*a*). Around 1518 hrs. phase path variations (record not shown) have indicated a perturbation first on *'x'* and later in *'o'* component. Again a second perturbation [indicated by the movement of fringes parallel to record length shown with an arrow marked *'p'* in Fig. 1 (*b*)] appeared only on *'x'* component at about 1522 hrs. and after that, phase path variations exhibited the oscillations in an unusual way with short period reversals. This is a new phenomenon, not noticed during earlier observations. Samples (*c*), (*d*), (*e*), (*f*) show continuous series of these oscillations. The sequence of these oscillations continued upto 1542 hrs. and finally disappeared with a pair of two reversals between 1543 hrs. and 1548 hrs. as shown in sample (*g*).

TABLE I

Details of the four events in which short period waves are observed on phase path records during magnetically disturbed days

Date of event	Time of occurrence in hours LT. (LT = UT + 5·5 hours)	Maximum value of 3 hourly magnetic indices occurred during or neighbouring interval of the event		Sum of magnetic indices of the day	
		K_p	K	K_p	K
22–3–1974	1355–1410	6	5	36+	30
18–4–1974	1330–1430	6	6	38+	31
19–4–1974	1520–1550	6	5	35+	30
21–4–1974	1400–1545	6	4	35−	27

K—Values obtained from the nearest Magnetic Observatory, Alibag (Geomagnetic 9·5° N).
K_p —Values published in the Solar Geophysical Data.

By observing the similarity of the corresponding reversals on *'o'* and *'x'* components in various samples of Fig. 1, it can be seen that they occur earlier on *'x'* component than *'o'* by a few seconds. Information from magnetic data (K_p values) indicated that 19–4–1974 was one of the five most magnetically disturbed days with a moderate activity all through the day and with a maximum K_p value at 6.

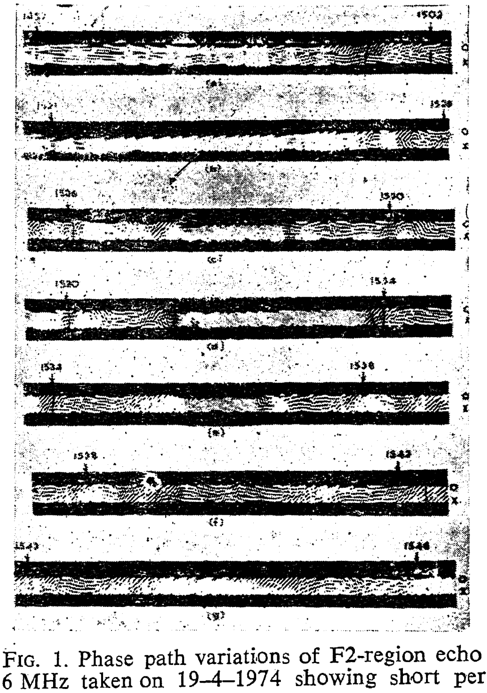

FIG. 1. Phase path variations of F2-region echo on 5·6 MHz taken on 19–4–1974 showing short period oscillations.

The events of these short period oscillations are seen in about four occasions. The details of these events and the maximum value of 3 hourly magnetic indices (which occurred either within the interval or neighbouring interval of the observed event) are presented in Table. I. From this table it can be inferred that the observed short period oscillations occurred consistently around 1400–1500 hrs. Their appearance is not noticed in the records obtained during the other part of the respective days given in Table I.

The time period between the midpoints of the corresponding reversals are measured and twice the value of this gives the period of the disturbance causing the oscillations at the reflecting height. The frequently observed periods are found to range between 50–80 seconds. This range of periods fall within the short period range of expected acoustic waves in the F-region (Fig. 9 of Georges[3]). For all of these events the occurrence of the disturbance is seen first on *'x'* component and later on *'o'*. These time delays range between 10–20 seconds and at certain parts of the record time, delays less than 10 seconds are also noticed. These low values of time delays suggest that the observed short period oscillations are caused by disturbances travelling faster than the normally observed TIDs[7] (Travelling Ionospheric Disturbances). This is a possibility in the case of acoustic waves. Hence it suggests that the observed short period oscillations are caused by the propagation of acoustic waves on these occasions.

From the available information there is no evidence to believe that the observed short period waves are caused by any source of the type mentioned by earlier workers such as tropospheric storms[2,3], earth quakes[1], nuclear detonations[4,5], etc. Hence it is felt that the only source associated with the observed oscillations are connected with the increased magnetic activity in view of the information indicated in Table I.

Existence of infrasonic waves at auroral latitudes at times of polar magnetic substorms is well understood[8]. Appearance of such waves at equatorial region is not noticed previously. From the observations discussed here one point to be stressed is that the occurrence of the events around local noon is in accordance with the occurrence of the geomagnetic activity effect on thermospheric density which is also maximum during local noon at the equatorial region at times of magnetic storms[9]. Possibility for the local generation of shorter period disturbances has been suggested by Hines[10] in explaining the greater speed disturbances at times of high magnetic activity. In view of this the present observations might be a representative one from a low latitude station like Waltair. However more number of similar observations from equatorial stations may lead to better understanding of these events.

Thanks are due to Dr. B. V. Krishna Murty of I.S.R.O. (Trivandrum) for useful discussions.

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1. Davies, K. and Baker, D. M., *J. Geophys. Res.*, 1965, 70, 2251.
2. — and Jones, J. E., *J. Atmos. Sci.*, 1971, 28, 254.
3. Georges, T. M., *J. Atmos. Terr. Phys:*, 1968, 30, 735.
4. Baker, D. M. and Davies, K., *J. Geophys. Res.*, 1968, 73, 448.
5. Barry, G. H., Griffiths, L. J. and Taenzer, J. C., *Ibid.*, 1966, 71, 4173.
6. Reddy, C. R. and Rao, B. R., *Radio Electron. Engr.*, July 1967, 34 (1), 53.
7. — and —, *J. Atmos. Terr. Phys.*, 1971, 33, 251.
8. Wilson, C. R., *Ibid.*, 1975, 37, 973.
9. Romer, M., *Space Res,,* 1970, 11, 965.
10. Hines, C. O., *Can. J. Phys.*, 1960, 38, 1441.

STUDIES ON THE CMC OF ANIONIC SURFACTANTS IN THE PRESENCE OF NONIONIC SURFACTANTS

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ABSTRACT

The effect of four non ionic surfactants, *viz.*, Tween 20 and 40, Nonidet P 40 and Nonex 501 on the CMC of three ionic surfactants, *viz.*, sulphonated phenyl stearic acid (SPSA), sulphonated tolyl stearic acid (STSA) and sulphonated xylyl stearic acid (SXSA) has been studied. Tha shift in the CMC of the ionic surfactants caused by the addition of nonionic surfactants has been interpreted on the basis of mixed micelle formation.

INTRODUCTION

SURFACE active agents are often mixed together to impart special properties (such as higher foaming power) to the resulting formulations, which find wide applications in industry[1]. The changes in the CMC of the resulting formulations give valuable information regarding their properties. A large amount of work has been carried out on the change of CMC of ionic surfactants as a function of the amounts of various additives. However, the effect of nonionic surfactants on the micellisation of anionic surfactants has received less attention. Studies have therefore been carried out on the effect of four nonionic surfactants, *viz.*, Tween 20 and 40, Nonidet P 40 and Nonex 501 on the CMC of three alkyl aryl sulphonates, *viz.*, sulphonated phenyl stearic acid (SPSA), sulphonated tolyl stearic acid (STSA) and sulphonated xylyl stearic acid (SXSA).

EXPERIMENTAL

The anionic surfactants, sulphonated phenyl-, tolyl-, and xylyl stearic acids were prepared by the method of Stirton *et al.*[2]. Nonionic surfactants, *viz*, Tween 20, Tween 40, Nonidet P 40 and Nonex 501 were BDH products. Surface tension *vs* concentration curves were plotted for each one of these surfactants as a criterion of purity. All the surfactants used showed sharp breaks in these plots which indicated their purity and homogeneity. The solutions of sulphonated aryl stearic acids (SPSA, STSA and SXSA) were prepared in double distilled water and their strengths determined by titrating pH metrically against standard KOH. The nonionic surfactants were dissolved in double distilled water and the solutions allowed to stand overnight to attain equilibrium.

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The CMC's of the anionic surfactants alone and in the presence of nonionic surfactants were determined by measuring specific conductance as a function of concentration with a Toshniwal conductivity bridge model CLOl/A using a dip type conductivity cell (cell constant = 0·74). It was used with bright platinum electrodes because detergents are srtongly absorbed from solution by spongy platinum. All measurements were carried out at 25 ± 1° C.

TABLE I

CMC values of ionic surfactants (SPSA, STSA, SXSA) in the presence of varying amounts of nonionic surfactants

Concentration of nonionic in g/dl	CMC × 10^{-4} M		
	SPSA	STSA	SXSA
0·0	3·00	3·25	3·60
Tween 20			
0·02	3·15	3·40	3·75
0·05	3·25	3·55	3·90
0·10	3·40	3·70	4·00
0·25	3·50	3·80	4·10
0·50	No inflection point		
Tween 40			
0·02	3·20	3·50	3·80
0·05	3·35	3·65	3·95
0·10	3·50	3·80	4·10
0·25	3·60	3·90	4·20
0·50	No inflection point		
Nonidet P 40			
0·02	3·10	3·45	3·70
0·05	3·30	3·60	3·80
0·10	3·45	3·75	3·90
0·25	3·55	3·85	4·00
0·50	No inflection point		
Nonex 501			
0·02	3·20	3·50	3·80
0·05	3·25	3·60	3·90
0·10	3·35	3·65	4·00
0·25	3·45	3·75	4·10
0·50	No inflection point		

CMC (by surface tension method) of: Tween 20—0·106 × 10^{-2} g/dl, Tween 40—0·071 × 10^{-2} g/dl, Nonidet P 40—0·12 × 10^{-2} g/dl, Nonex 501—0·062 × 10^{-2} g/dl.

RESULTS AND DISCUSSION

The plots of specific conductance *versus* concentration of the anionic surfactant show inflections, which correspond to their critical micelle concentration (CMC). It is observed that the CMC occurs at progressively higher concentration of the anionic surfactant on increasing the concentration of the added nonionic surfactant (Table I). It is further observed that the inflections become increasingly less pronounced at higher concentration of the nonionic surfactant and ultimately disappears when the concentration reaches 0·5 g/dl. The effect of nonionics on increasing the CMC of the anionics may be traced to their effect on the so-called iceberg structure in the water[3,4] (breaking of the water structure) as originally proposed by Frank and Evans[5] and the formation of mixed micelles[6-9]. The factors responsible for the formation of mixed micelles in the mixtures of anionic-nonionic surfactants appear to be : (*i*) Van der Waals attraction between the hydrophobic chains, (*ii*) hydrogen bonding through the ether oxygens of the polyoxethylene chains of the nonionic species[7,10 11] and (*iii*) electrostatic effects, since the nonionic surfactants are not truly nonionic but possess a small positive charge arising from axonium ion formation[12]. The disappearance of the inflection point at a concentration of 0·5 g/dl of the nonionic may be attributed to the ability of a single ethylene oxide chain to solubilize many ionic groups. Therefore, beyond this concentration of the nonionic, the anionics seem to always exist as mixed micelles and hence no transition point is possible[10,13].

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1. Schwarz, A. M. and Perry, W. J., *Surface Active Agents,* Interscience, New York, Vol. 1, 1949, p. 236.
2. Stirton, A. J., Peterson, R. F. and Gorggins, P. H., *Ind. Eng. Chem.,* 1940, 32.
3. Malik W. U. and Verma, S. P., *Kolloid Z.,* 1969, 233, 985.
4. — and Jhamb, O. P., *Ibid..* 1970, 242, 1209.
5. Frank, H. S. and Evans, M. W., *J. Chem. Phys.,* 1945, 13, 507.
6. Yoda, O., Miguro, K., Kondo, T. and Groggins, P. H., *Ind. Eng. Chem.,* 1940, 32, 1136.
7. Shinoda, K. and Hildebrand, J. H., *J. Phys. Chem.,* 1957, 61, 789.
8. Makagawa, T. and Inova, H.. *J. Chem. Soc. Japan, Pure Chem. seet..* 1957, 78, 636.
9. Corkill, J. M., Goodman, J. F. and Tate, J. R., *Trans. Faraday Soc.,* 1964, 60, 986.
10. Schick, M. J. and Manning, D. J., *J. Am. Oil Chem. Soc..* 1966. 43, 133.
11. Malcomn. N. J., *J. Colloid Sci.,* 1967, 23, 36.
12. Bembi. R.. Goyal, R. N. and Malik, W. U., *Talanta* (in press).
13. Schick. M. J., *Nonionic Surfactants,* Marcel Dekker, New York, Vol. 1, 1967, p. 508.

LETTERS TO THE EDITOR

SOLAR CYCLE CONSTANT OF IONOSPHERIC ABSORPTION

THE solar cycle variation of ionospheric absorption of pulsed radio transmissions at 2·4 MHz measured at Waltair during the period 1971–73 has been studied to obtain the solar cycle constant *'b'* using the relation $L = L_0 (1 + bR)$ in which L is the noon absorption in dB, R is the Zurich sunspot number and L_0 is the absorption in dB for $R = 0$. Earlier measurements of solar cycle constant *'b'* at equatorial and low latitudes showed wide variability[1-4]. Hunter[5] drew attention to this discrepancy in the measured value of *'b'* and suggested further measurements. Subsequently Mbipom[6] re-examined the ionospheric data at Ibadan with additional data on 2·4 MHz echoes obtained during 1966–68. He found that *'b'* values are in close agreement at Colombo and Ibadan. More recently Kotadia *et al.*[7] studied the solar cycle variation of absorption on 2·2 and 2·5 MHz at Ahmedabad. The availability of absorption data on 2·4 MHz enabled us to study the solar cycle variation and to see whether the result is consistent with those at other equatorial stations.

TABLE I

Monthly mean values of 'b' *in* $L = L\ (1 + bR)$

	Jan.	Feb.	Mar.	April	May	June	July	Aug.	Sept.	Oct.	Nov.	Dec.	Mean
$b \times 10^3$	5·7	4·5	4·7	4·5	8·1	5·1	5·7	6·6	9·4	5·3	6·4	6·9	6·1
L_0 (dB)	35	44	42	44	37	39	35	30	32	38	31	29	36

The seasonal control on absorption has to be removed to study the sunspot cycle variation. It is done in two ways, either by taking 12 month running means or by studying the variation for each individual month. The latter method was used by Appleton and Piggott[8] and Lastovicka[9] and is useful since it gives the parameter *'b'* for each month. In this method the absorption data is grouped month-wise and plotted against Zurich sunspot number (obtained from solar geophysical data bulletins) and best fit lines are drawn. The slopes of these best fit lines give the values of *'b'* and the average values are given in Table I. The monthly mean values of the solar cycle constant *'b'* vary from a minimum of $4·5 \times 10^{-3}$ to a maximum of $9·4 \times 10^{-3}$. The annual mean value of *'b'* obtained is $6·1 \times 10^{-3}$. If the two high values of *'b'* obtained in May and September are omitted, the annual mean becomes $5·5 \times 10^{-3}$. It may not be out of place here to mention that the monthly mean *'b'* values, derived from Al absorption data by Appleton and Piggott[8] showed wide variation from month to month while the *'b'* values derived from f_oE data were practically uniform throughout the year. The variation in the monthly mean values of *'b'* is mainly due to the different contributions of deviative absorption to the total absorption.

It is clear from Table II, in which the average value of *'b'* obtained in the present investigation is compared with those at other equatorial stations, that our value of *'b'* is reasonably in good agreement with those at other stations and the discrepancy observed in earlier measurements are no longer evident. One important conclusion from this study is that the solar cycle constant at low and equatorial latitudes and for E region echoes can be taken as $5·5 \times 10^{-3}$ approximately.

TABLE II

Values of solar cycle constant 'b' *at different low latitude stations*

Station	Frequency MHz	Period of observation	$b \times 10^3$	Ref. No.
Colombo	2·6	1964–68	5·1	4
Ibadan	2·4	1955–68	4·9	6
Singapore	2·4	1954–58	4·8	4
Ahmedabad	2·2	1972–75	6·6	7
Ahmedabad	2·5	1972–75	5·0	7
Waltair	2·4	1971–73	6·1	Present work

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---

1. Shirke, J. S., *J. Telecomm. Engrs.*, 1961, **5**, 115.
2. Skinner, N. J. and Wright, R. W. H., *J. Atmosph. Terr. Phys.*, 1964, **26**, 1221.
3. Rao, A. S M., *Ph.D. Thesis,* Andhra University, Waltair, 1967.
4. Gnanalingam, S., *Proc. III Int. Symp. on Equatorial Aeronomy,* PRL, Ahmedabad, 1969.
5. Hunter, A. N., *Proc. II Int. Symp. on Equatorial Aeronomy* (Ed. F. De Mendōnca).
6. Mbipom, E. W., *J. Atmosph. Terr. Phys.*, 1971, **32**, 1263.
7. Kotadia, K. M., Gupta, A. and Chipa, G. M., *Paper presented at the Symp. on Ionosphere and Magnetosphere,* Andhra University, Waltair, Jan. 1976 (Abstract).
8. Appleton, E. V. and Piggott, W. R., *J. Atmosph. Terr. Phys.,* 1954, **5**, 141.
9. Lastovicka, J., *Geophysicalni Sbornik,* 1969, p. 291.

ON THE L-CONVERSION OF THE 77 keV TRANSITION IN ^{197}Au

THE radioactive isotope ^{197}Hg essentially decays (98%) to the 77 keV state in ^{197}Au, which subsequently decays to the ground state (3/2+). The nature of the 77 keV transition is of interest from the viewpoint of inferring the structure of the 77 keV state. The transition properties are summarized in the recent *Nuclear Data Sheets*[1]. On the basis of the Deshalit's model[2], the 77 keV state was essentially considered as a core excitation state. On the other hand in the wave functions furnished by Kisslinger and Sorensen[3], a large part of this state was viewed as of single particle origin. The state was also considered on the basis of the Intermediate coupling model in the Unified Scheme with considerable success[4]. Since the energy of the state is lower than the K-shell binding energy in Au (B_k = 80·7), K-conversion does not take place. In the present experiment, the L-shell conversion coefficient of the 77 keV transition is determined since no such determination was made earlier[1], although L-subshell ratios were studied.

A direct determination of the L-conversion coefficient is possible from the L-X-ray and the 77 keV gamma ray intensities recorded with a Si (Li) detector. This method, however, is not used in the present study since it is hard to correct from the contributions of other transitions to the L-X-ray. In addition, the 77 keV gamma ray is not resolved completely from the K$_\beta$ X-ray line arising due to the K-conversion of other transitions. Hence a coincidence method is employed for the determination of the L-conversion coefficient in the present study, using a Ge-(Li)-NaI (Tl) coincidence system with a ND 512 channel analyser. The Ge (Li) detector (coaxial 35 cc) is employed to detect the K-X-rays associated with the K-capture, while the NaI (Tl) detector (1½″ dia × 1 mm thick with a thin aluminium window, attached to DuMont 6292 photomultiplier), is employed to detect the X-rays following L-capture and gamma rays associated with the 77 keV transition. The output from the Ge (Li) detector is employed to gate the multichannel analyzer in which the coincident spectrum detected in the NaI (Tl) is recorded. The resultant spectrum shows peaks corresponding to the 77 keV gamma ray and the L-X-ray. The intensities under these lines N$_L$ and N$_\gamma$ obtained by graphical analysis are used to estimate the L-shell conversion coefficient using the relation

$$\alpha_L\ (77\ \mathrm{keV}) = \frac{N_L}{N_\gamma} \cdot \frac{\epsilon_\gamma}{\epsilon_L} \cdot \frac{1}{\omega_L} \qquad (1)$$

where ϵ_γ and ϵ_L are the relative photopeak efficiences and ω is the L-shell fluorescent yield which is taken as — 0·41 ± 0·04 from the data of Bambynek *et al.*[5]. An auxiliary experiment was conducted to determine the relative intensities using a ^{57}Co source which yielded $\epsilon_\gamma / \epsilon_L = 0{\cdot}96 \pm 0{\cdot}03$. The final value of the L-conversion coefficient is obtained as

$$\alpha_L\ (77\ \mathrm{keV}) = 2{\cdot}46 \pm 0{\cdot}26$$

which includes an overall error of 10%, essentially due to the error in the L-shell fluorescent yields. The statistics of counting did not involve more than 1% error. The theoretical values obtained from the computer interpolation programme of Hager and Seltzer[6] are

$$\alpha_L\ (M\,1) = 2{\cdot}28$$
$$\alpha_L\ (E\,2) = 12{\cdot}8,$$

The present value of α_L suggests an essentially M 1 assignment to the 77 keV transitoin. Young *et al.*,[7] obtained a value for the total conversion coefficient from their coulomb excitation study as 4·0 ± 0·2 and employed it for the estimation of the penetration parameter. Assuming this penetration parameter and the value of the mixing ratio derived from Mossbauer study (δ^2 = 8·1218 ± 0·0007), L-conversion coefficient is estimated with the theoretical 'no penetration' values. The resultant value is obtained as,

$$\alpha_L\ (77\ \mathrm{keV})\ \mathrm{Young} = 3{\cdot}12 \pm 0{\cdot}10.$$

A similar estimate, made from the data of Krpic *et al.*[8] yielded α_L (77 keV) (Krpic) = 2·92 ± 0·3.

The present result is closer to that of Krpic *et al.*, which in turn means that the present value indicates a different penetration parameter. Using the present experimental value for the L-shell conversion coefficient and the theoretical 'no penetration' values, the penetration parameter is estimated to be $\lambda = 11 \pm 3$. The value is not inconsistent with the systematics of Subba Rao[9], who fitted the penetration parameters in an empirical relation with M 1 retardation factors.

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---

1. Lewis, M. B., *Nuclear Data Sheets*, 1972, 7 **B**, 129.
2. De Shalit, A., *Phy. Letters*, 1965, 12, 170; Braustein, A. and De Shalit, A., *Ibid.*, 1962, 1, 264.
3. Kisslinger, L. S. and Sorensen, R. A., *Rev. of Mod. Phys.*, 1963, 35, 853.
4. Ila, P., *Ph.D. Thesis*, Andhra University, 1974.
5. Barnbynek, W., Crasamann, B., Fink, R. W., Freund, H. U., Mark, H., Swift, C. D., Price, R. E. and Venugopala Rao, P., *Rev. Mod. Phys.*, 1972, 44, 716.
6. Hager, R. S. and Seltzer, E. C., *Nuclear Data Sheets*, 1968, 4 **A**, 1.
7. Young, L. E., Brennand, R. and Sprouse, G. D., *Nucl. Phys.*, 1974, 229 **A**, 157.
8. Krpic, D., Stepic, R. Bikit, I., Milanovic, J. and Vukanovic, R., *Z. Phy.*, 1971, 243, 452.
9. Subba Rao, B. N., *Phy. Letters*, 1975, 56 **B**, 435.

ON THE CONVERSION OF THE 192 keV TRANSITION IN ^{114}In

The 192 KeV transition in the decay of the isomeric state ^{114m}In, is exetnsively used as a standard gamma ray for energy and efficiency calibration of NaI(Tl) and Ge(Li) detectors. The most widely referred to conversion work on this transition is that of Hoffmann[1] who employed an anthrasene split crystal and obtained using peak-to-beta counting method $\alpha_T = 4 \cdot 3 \pm 0 \cdot 4$ and $\alpha_k = 2 \cdot 8 \pm 0 \cdot 5$. The latter value was extracted using the K/L vaule of $2 \cdot 0 \pm 0 \cdot 6$ The values of K/L and K/L + M obtained by other authors[2] were at variance with the value obtained by Hoffmann, values of 1·12 to 2·0 being reported. A careful measurement of the K-conversion coefficient is therefore carried out in the present study. In view of the simple decay scheme associated with this isotope, K X-ray-to-gamma ray method is applicable in this case. However, a Ge(Li) spectrometer system is employed to separate out the interfering events from the genuine ones.

It is well established[2] that ^{114m}In decays by the isomeric transition in 96·6% of the cases and by electron capture to levels in ^{114}Cd in the remaining 3·4% of the cases. The ground state decay of the ^{114}In is mostly associated with the negatron decay to ^{114}Cd. The nature of the 192 keV isomeric transition is established to be E4, the connecting state characteristics being well established to be 5^+ and 1^+. The internal conversion of the 192 keV transition in ^{114}In gives rise to In K X-rays while the electron capture decay contributes to the X-rays of Cd. However, since the energy difference of these X-rays is small, they are counted together. The total contribution of the X-rays through the electron capture is estimated employing the percentage branches, the K-capture probability P_k, the internal conversion associated with the gamma rays associated with Cd and the fluorescent yield of Cd. The K-capture probability is estimated using the relations furnished by Martin *et al.*[3]. The K-conversion coefficients are obtained from literature[4] and the fluorescent yields are obtained from the data of Bambynek *et al.*[5]. The total contribution to the X-ray peak thus estimated is found to be 6%, the remaining 94% being due to the conversion of the 192 keV transition. Since the contribution is small, the error associated with the contribution could be neglected. Thus, after correcting for the small other contributions, the K X-ray intensity could be used together with the gamma ray intensity to evaluate the K-conversion coefficient using the relation

$$\alpha_k \text{ (192 keV)} = \frac{N_x}{N_\gamma} \cdot \frac{\epsilon_\gamma}{\epsilon_x} \cdot \frac{1}{\omega_k} \tag{1}$$

where N's are the intensities, ϵ's are the relative photopeak efficiences and ω_k is the K-shell fluorescent yield in In.

The radioactive source ^{114m}In is obtained from the Isotope Division, BARC as $InCl_3$ in HCl solution and a few drops of the source liquid dried over a mylar foil over a perspex frame are employed for the determination of the conversion coefficient. The source is directly mounted on the cryostat arm of the 35 cc coaxial Ge(Li) detector and the spectrum is recorded on a ND 512 channel analyzer. The relative efficiency calibration is carried out using auxiliary sources ^{152}Eu, ^{133}Ba, ^{75}Se and ^{57}Co. Using the intensities N_x and N_γ under the X-ray and gamma lines together with the relative efficiencies and $\omega_k = 0\cdot850 \pm 0\cdot029$ taken from the data of Bambynek *et al.*,[5] the final value of α_k is obtained as

$$\alpha_k \text{ (192 keV)} = 2\cdot00 \pm 00\cdot09$$

The error is mostly contributed by that in the fluorescent yield, the error in the statistics of counting being only of the order of 1%. The present value is more accurate than the value reported in the Nuclear Data Sheets. The theoretical value obtained from the computer interpolation programme of Hagar and Seltzer[6] for a 191·6 keV transition of E4 type is 2·48. The present value of the experimental conversion coefficient is smaller than the theoretical value by (19 ± 4)%. It may be recorded that Raman *et al.*[7] concluded in their survey that the conversion coefficients of high multipole transitions (E3, M4) to show discrepancies between theory and experiment. The present transition which is an E4 follows the same trend, but it is of interest to note that deviation is much larger. The gamma ray transition probability is found to show a hindrance of about 30 over the single particle estimate (Moszkowiski), in correlation with the present anomalous conversion coefficient. This is indicative of the differences in the structure of the connecting states.

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---

1. Hoffmann, K. W., *Z. Physik*, 1957, 148, 298.
2. Kim, H. J., *Nuclear Data Sheets*, 1975, 16, 107.
3. Martin, M. J. and Blichert-Toft, P. H., *Ibid.*, 1970, 8A, 157.
4. Hamilton, J. H., *Rev. of Experimental and Theoretical Conversion Coefficients* (Private Communication), 1974.
5. Bambynek, W., Crasemann, B., Fink, R. W., Freund, H. U., Mark, H., Swift, C. D., Price, R. E. and Venugopala Rao, P., *Rev. Mod. Physics*, 1972, 44, 716.
6. Hager, R. S. and Seltzer, E. C., *Nucl. Data Tables*, 1968, 4A, 1.
7. Raman, S., Walkiewicz, T. A., Gunnik, R. and Martin, B., *Phys. Rev.* 1973, 7C, 2531.

THE CRYSTAL STRUCTURE OF TRIGLYCINE CALCIUM BROMIDE

THE crystal structure of diglycine calcium chloride tetrahydrate was already reported[1] and the structure determination of the addition compound of glycine with calcium iodide is under progress. This is a preliminary note reporting the atomic coordinates in the crystal structure of triglycine calcium bromide.

The single crystals of triglycine calcium bromide [$(NH_2CH_2COOH)_3CaBr_2$] were grown from a saturated aqueous solution containing glycine and calcium bromide. The crystal data are as follows: $a = 8\cdot97$, $b = 14\cdot74$, $c = 20\cdot30$ Å, $V = 2681\cdot5$ Å^3, F. W = 425·1 gm., $D_{mea} = 2\cdot06$ gm. cm^{-3}, $D_{cal} = 2\cdot10$ gm. cm^{-3}, $Z = 8$, μ (CuK_α) = 114·7 cm^{-1} and the space group is $Pbc2_1$ which is confirmed by structure analysis. The density was measured by flotation, in a mixture of bromoform and carbon tetrachloride.

The three-dimensional intensity data (Hkl, $H = 0$ to 8, $\mu r \approx 1\cdot02$; hKl, $K = 0$ and 1, $\mu r = 1\cdot21$) were collected using CuK_α radiation by the multiple-film equi-inclination Weissenberg technique. The spot-shape, absorption, Lorentz and polarisation corrections were applied in the usual way. Peaks with coordinates (0, 0, w) were absent in the Patterson synthesis indicating that the space group is non-centric. From sharpened and unsharpened three-dimensional Patterson syntheses, as well as from a three-dimensional minimum function, the positions of the four bromine atoms and the two calcium atoms in the asymmetric part were determined.

Thereafter, successive Fourier and difference Fourier syntheses revealed the rest of the structure. With isotropic thermal parameters for all the atoms, least-squares refinement was carried out using the LALS program (originally written by Gantzel, Sparks and Trueblood and modified later by Liminga of Uppsala and also in the University of Madras) on the IBM 370/155 computer, at the Indian Institute of Technology, Madras. For 1576 observed reflections, the residual index at present is 0·13. Further refinement is under progress. The present fractional atomic coordinates are recorded in Table I.

Each of the calcium atoms is coordinated to five oxygen atoms of the carboxyl groups and a bromine atom. The Ca-O distances range from 2·01 to 2·72 Å, and the Ca-Br distances are 2·93 Å and 3·02 Å. The glycine molecules have the normal bond distances and bond angles as found in other similar structures[2]. The hydrogen bonds are between the nitrogen, oxygen and bromine atoms.

This compound was earlier assigned the chemical formula $(NH_2CH_2COOH)_2CaBr_2\cdot4\,H_2O$, similar to that of the diglycine calcium chloride tetrahydrate[1]. But, the present structure analysis undoubtedly confirms the chemical formula $(NH_2CH_2COOH)_3CaBr_2$, the molecular weight of which is more or less the same as that of diglycine calcium bromide tetrahydrate.

TABLE I

Fractional atomic coordinates for triglycine calcium bromide

Atom	x	y	z
Br (1)	−0·2556	0·1778	0·3556
Br (2)	0·2722	0·2056	0·3750
Br (3)	0·2278	0·4222	0·2444
Br (4)	−0·2444	0·4611	0·2416
Ca (1)	0·1750	0·2361	0·0111
Ca (2)	0·3278	0·0139	0·1028
O (11)	0·177	0·139	0·121
O (21)	0·025	0·258	0·110
C (11)	0·083	0·194	0·142
C (21)	0·045	0·183	0·215
N (11)	−0·058	0·269	0·253
O (12)	0·322	0·110	−0·002
O (22)	0·472	−0·011	0·000
C (12)	0·411	0·056	−0·028
C (22)	0·456	0·069	−0·100
N (12)	0·533	−0·011	−0·131
O (13)	0·069	0·383	0·000
O (23)	−0·128	0·443	0·052
C (13)	0·011	0·433	0·044
C (23)	0·097	0·494	0·092
N (13)	0·260	0·488	0·076
O (14)	0·369	0·311	0·061
O (24)	0·571	0·376	0·108
C (14)	0·508	0·322	0·069
C (24)	0·596	0·263	0·020
N (14)	0·760	0·258	0·034
O (15)	−0·198	0·064	−0·056
O (25)	−0·042	0·158	−0·028
C (15)	−0·069	0·094	−0·064
C (25)	0·039	0·047	−0·111
N (15)	−0·037	−0·035	−0·139
O (16)	0·708	0·192	0·167
O (26)	0·528	0·094	0·133
C (16)	0·583	0·156	0·167
C (26)	0·467	0·206	0·211
N (16)	0·525	0·291	0·242

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---

1. Natarajan, S. and Mohana Rao, J. K., *Curr. Sci.*, 1976, **45**, 490.
2. Mohana Rao, J. K. and Viswamitra, M. A., *Acta Cryst.*, 1972, **28 B**, 1484.

POTASSIUM BROMATE OXIDATION OF ARYL METHYL SULPHIDES

NUMEROUS oxidizing agents[1,2] have been used for the transformation of dialkyl, diaryl and alkyl aryl sulphides to sulphoxides. The products are usually contaminated with the sulphides and/or the corresponding sulphones. Good yields of sulphoxides are reported in oxidations using potassium periodate[2], *t*-butyl hypochlorite[3], N-halosuccinimides[4] and iodobenzene dichloride[4a], but a low temperature is required for the reaction.

We have now observed that the reaction between aryl methyl sulphides and potassium bromate in 3 : 1 mole ratio at room temperature yields sulphoxides in high yield and purity. The sulphoxides have been characterised by elemental analysis, boiling and/or melting point and mixed melting point (Table I). Their IR and UV absorption spectra are identical with those of the authentic samples obtained by chromic acid[5] oxidation of the sulphides. A very strong S=O stretch absorption occurs in the 1035–1025 cm^{-1} region and the UV absorption maximum occurs around 238 nm in ethanol for these sulphoxides. These values agree with those reported in the literature[6,7].

The reaction in 1 : 1 mole ratio at 100° C leads to the formation of sulphones in 60–70% yield, which are characterised by melting points and mixed melting points with authentic samples (Table II). This study demonstrates a convenient method to obtain sulphoxides in high yield and purity.

Experimental

1. *Aryl methyl sulphoxides*: *General method.*—To a solution of aryl methyl sulphide (0·051 mole) in glacial acetic acid (25 ml) was added Analar potassium bromate (0·017 mole) dissolved in minimum amount of water, in small portions with shaking and cooling under the tap. The mixture was strongly coloured. If any turbidity developed more glacial acetic acid was added to make it homogeneous and then it was allowed to stand for an hour. It was poured into 200 ml of water and neutralised with sodium carbonate. The organic layer was extracted with chloroform and dried over anhydrous sodium sulphate. The solvent was removed under reduced pressure and the liquid distilled under vaccum.

In the case of *p*-bromophenylmethyl sulphide the sulphoxide separated as soon as the addition of potassium bromate was completed. It was collected on a filter and crystallisation from aqueous ethanol afforded colourless plates.

2. *Aryl methyl sulphones* : *General method.*—Potassium bromate (0·03 mole) dissolved in the minimum quantity of hot water was added to a hot

TABLE I

Characterization data for sulphoxides Ar–SO–CH₃

Ar	Yield, %	B.P./mm, (M.P.), °C		Reference	S=O (ν cm^{-1})*
		Found	Literature		
C_6H_5	80	118–20/5	144/15	8	1030
p-$CH_3C_6H_4$	86	138–40/12 (42)	113–4/2 (42–43)	8	1030
o-$CH_3C_6H_4$	85	138–9/9·5	..	..	1032
p-ClC_6H_4	86	135–6/5 (48)	135–6/5 (47–48)	8	1028
p-BrC_6H_4	95	(87)	(86–87)	9	1033**
p-$CH_3OC_6H_4$	75	152–3/5	153–4/5	8	1035
o-$CH_3OC_6H_4$	77	148–50/5	..	..	1025

* IR spectra were recorded on Perkin-Elmer 257 (neat).
** IR spectra in KBr pellet.

TABLE II

Ar-SO₂-CH₃

Ar	Yield, %	M.P., °C		Reference
		Found	Literature	
C_6H_5	60	88	88	1
p-$CH_3C_6H_4$	65	88–89	89	11
p-ClC_6H_4	62	96	96	12
p-BrC_6H_4	70	99	99–100	9

solution of the aryl methyl sulphide (0·03 mole) in glacial acetic acid (25 ml), kept on a water-bath and allowed to stand for an hour. It was cooled, poured into crushed ice, stirred and the pricipitated sulphone was collected on a filter and recrystallised from suitable solvents.

Kinetics of oxidation to sulphoxide are under investigation.

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---

1. Reid, E., *Organic Chemistry of Bivalent Sulphur,* Vol. II, Chemical Publishing Co., Inc., New York, 1960, p. 64.
2. Leonard, N. J. and Jhonson, C. R., *J. Org. Chem.,* 1962, 27, 283.
3. (*a*) Walling C. and Mintz, M. J., *Ibid.,* 1967, 32, 1286.
   (*b*) Skattebol, L., Boulette, B. and Solomon, S., *Ibid.,* 1967, 32, 3111.
4. Harville, R. and Reed Jr., S. F., *Ibid.,* 1968, 33, 3976.
   (*a*) Barbieri, G. *et al., J. Chem. Soc (C),* 1968, p. 659.
5. Edwards, D. and Stenlake, J. B., *J. Chem. Soc.,* 1954, p. 3272.
6. Bellamy, E. J., *The Infra-red Spectra of Complex Molecules,* John Wiley and Sons, Inc., New York, 1958, p. 358.
7. Leandri, G., Mangini, A. and Passerini, R., *J. Chem. Soc.,* 1957, p. 1386.
8. Modena, G., *Gazz. Chim. Ital.,* 1959, **89**, 843.
9. Sergeer, P. G., *J. Gen. Chem.* (*USSR*), 1931, 1, 279 ; *C.A.,* 1931, 26, 2184.
10. Bohme, H. and Fischer, H., *Ber.,* 1942, 75, 1310.
11. Zincke, Th. and Frohneberg, W., *Ibid.,* 1910, 43, 848.
12. Miller, C. J. and Samuel Smiles, *J. Chem. Soc.,* 1925, 127, 224.

CATALYTIC DECOMPOSITION OF ISOPROPYL ALCOHOL ON Mg $Al_{2-x}Fe_xO_4$

THE catalytic properties of transition metal ions (Ni, Cr) in the environment of a diamagnetic matrix ($MgAl_2O_4$) has attracted a lot of attention in recent years[1,2]. Reports on the catalytic activity of the system Mg $Al_{2-x}Fe_xO_4$, are not available although this system shows interesting variation in the electrical and magnetic properties as x varies from 0 to 2[3,4]. This system is also interesting structurally, as while $MgAl_2O_4$ is a normal spinel, the end member $MgFe_2O_4$ with $x = 2$, is an inverse spinel. The present investigation is a part of our earlier studies on the catalytic activity of spinels[5].

The catalysts have been prepared by the co-precipitation of hydroxides of Mg, Al and Fe at an optimum pH (9·5) from the respective nitrates. The completion of precipitation of each ion was checked independently. The pricipitates were dried at 100° C and calcined at 950° C for 24 hours. The solid solutions formed were characterized by TGA, DTA and chemical analyses as well as by X-ray diffraction patterns. Isopropyl alcohol (BDH AnalaR) was distilled before use and its purity checked by using a chromatograph.

The decomposition of isopropyl alcohol was followed in a fixed bed flow type integral reactor and the liquid and gaseous products were analysed by chromatograph (Varian 1800), and gas analysis techniques respectively. The reaction was studied in the temperature range 300–370° C. From the plots of contact time *versus* concentration, the initial rates were computed and used for making the Arrhenius plots.

Analysis of the dehydration and dehydrogenation reaction products showed that when x has values of 0 or 0·02, the system exhibited only dehydration activity to the complete exclusion of dehydrogenation, although traces of the condensation product, namely, di-isopropyl-ether ($< 0\cdot5$ mole %) was also noticed. All the other catalysts exhibited both dehydration and dehydrogenation activities, though with varying selectivities. Fig. 1 shows the variation of selectivity for dehydration activity (s) at 350° C with change in titration metal ion (t.m.i) concentration. Selectivity was calculated as the ratio of the concentration of dehydration product to total decomposition. The selectivity reaches a minimum at $x = 0\cdot4$ and then picks up to a value of 25% at $x = 0\cdot667$ and afterwards remains practically the same though it drops down a bit in the case of pure $MgFe_2O_4$.

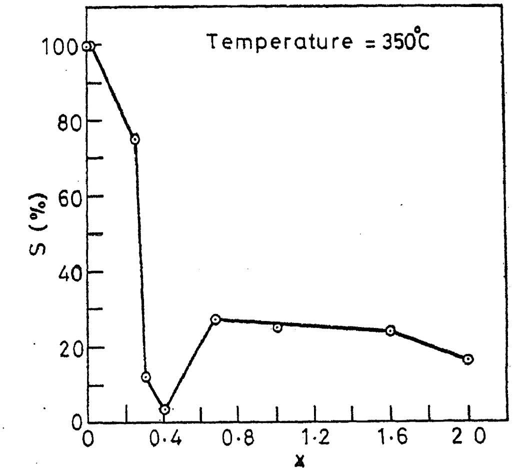

FIG. 1. Selectivity for Dehydration of Isopropyl Alcohol *vs.* x plot on $MgAl_{2-x}Fe_xO_4$ ($x = 0 \rightarrow 2$).

Arrhenius plots for the dehydration reaction for the various samples showed that for all catalysts with $x < 0\cdot4$, the activation energy values lie in the range of 26–32 kcal/mole, while the corresponding activation energies for the catalysts with $x > 0\cdot4$ are in the range of 8–15 kcal/mole. The activation energy for the catalyst with $x = 0\cdot4$ could not be obtained as the dehydration product was negligible and hence rates could not be calculated with accuracy.

From the activation energy values mentioned above, it may be postulated that the active sites for dehydration could be different on either side of minimum (Fig. 1) changing presumably from a surface coordinatively unsaturated Al^{3+} ions as in Al_2O_3[6] to Fe^{3+} ions as in Fe_2O_3 which are relatively weak Lewis acid sites. This postulate gains further strength from the fact that, whereas, water inhibits greatly the dehydration activity for catalysts with $x < 0\cdot4$, it does not have measurable effect on dehydration activity for catalysts with $x > 0\cdot4$. The change-over of selectivity towards predominant dehydogenation in the case of the catalysts with high concentration of iron could be attributed to predominant n-type semiconductivity of these catalysts, as revealed by our conductivity studies.

Any further comparison of dehydrogenation activities of the catalysts should take into account factors like the magnetic behaviour and the distribution of t.m.i in the matrix $MgAl_2O_4$ and these studies are in progress.

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Madras 600 036, *June* 1, 1976.

---

1. Cimino, A. and Schiavello, M., *J. Catal.*, 1971, 20,, 202.
2. Egerton, T. A. and Vickerman, J. C., *Ibid.*, 1970, 19, 74.
3. Kwestroo, W., *J. Inorg. Nucl. Chem.*, 1959, 9, 65.
4. Blasse, G. Philips, *Res. Rpt. Suppl.* 1964, 3, 1.
5. Narasimhan, C. S. and Swamy, C. S., *Curr. Sci.*, (in press) .
6. Krylov, O. V., *Catalysis by Nonmetals*, Academic Press, p. 131.

TRITERPENOIDS AND PHTHALYL ESTERS OF *MOGHANIA MACROPHYLLA* STEM-BARK

Moghania macrophylla Willd (Syn. *Flemingia macrophylla* Willd, *F. congesta* Roxb.; Leguminosae) is an erect shrub occurring at lower elevations throughout India and in Andaman Islands[1]. The earlier work on the mixture of leaves and flowers revealed the presence of several chromenochalkones[2]. The results of an examination of the stem-bark, obtained from Indian Lac Research Institute, Ranchi, Bihar, are described in this note.

The brown coloured, air dried and powdered stem-bark (790 gm) was exhaustively extracted with petrol (60–80°), benzene, acetone and methanol successively in a Soxhlet-extractor. The petroleum ether extract (4·3 gm) yielded an oily concentrate which was subjected to column chromatography over silica gel employing petroleum ether-benzene combinations and the following compounds were obtained in the order of elution and identified by direct comparison with authentic specimens and by the preparation of acetates : lupeol (40 mg), α-amyrin (30 mg) and sitosterol (50 mg).

Benzene and acetone extracts behaved similarly on T.L.C. and were combined. The oily residue was chromatographed over silica gel. Elution with light petrol-benzene (4 : 1) yielded a pinkish yellow syrupy liquid (1·73 gm) whose infrared spectrum showed adsorptions at 1720, 1280, 1120 (aromatic esters), 1600, 1580, 1312, 1280, 1070, 1035 and 740 cm^{-1} (ortho substituted benzene ring). 60 MH_z PMR spectrum (CCl_4) showed a complex but symmetrical multiplet for four protons at δ 7·58 (characteristic of AA' XX" type), two broad signals each for two protons at δ 4·20 and 4·30 (methylenes carrying the oxygen function) and aliphatic protons δ 0·80–1·70. Although it appeared to be homogeneous on TLC system [chloroform-methanol-water, (13 : 7 : 5) lower phase], its mass spectrum showed the presence of two peaks at m/e 278 and m/e 306 in roughly equal intensities. The presence of peaks characteristic of phthalyl esters at m/e 167 and m/e 149 of which the latter was the base peak, coupled with the generation of phthalic acid (m.p., m.m.p., Co-IR) on saponification suggested that the fraction is most likely a 1 : 1 mixture of dibutyl and diamyl phthalates. Further, mass spectra were not indicative of the presence of alkyl chain greater than C_5H_{11} unit.

Methanol extract: The brown coloured semi-solid (5·5 gm), obtained on concentrating the methanol extract under reduced pressure, gave positive test for proanthocyanidins and on several repetitions of the process of dissolution in methanol and precipitation by ether afforded light brown solid. This on hydrolysis with alcoholic 5 NHCl gave cyanidin (λ_{max} and paper chromatography) indicating the proanthocyanidin to be cyanidin based.

The ether soluble portion on column chromatography over silica gel yielded *sitosterol-β-D-glucoside* in ethyl acetate eluates (45 mg), m.p. 295–303° (d), $[\alpha]_D$ − 46·3° (pyridine). It gave positive LB, TNM and Molisch tests and the identity was confirmed by direct comparison (mixed m.p., Co-TLC and Co-IR); and by conversion to tetra acetate, m.p. 165–167°, $[\alpha]_D$ − 27·6° (*c*, 0·67 in $CHCl_3$).

The presence of alkyl phthalates has been recorded in some Umbelliferous plants[3,4], Balanitaceae[5] and Anacardiaceae[6]; this, however, is the first report of their occurrence in Leguminosae. As these compounds are known to be, at times, notorious contaminants from extraneous sources, their presence was confirmed by separate extractions of the bark avoiding contact with plastic and using phthalate free solvents. The presence of phthalyl ester in this species may be of chemotaxonomic significance.

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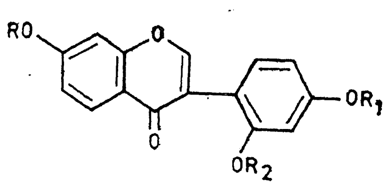
---

1 *Wealth of India, Raw Materials*, C.S.I.R., New Delhi, 1956, 6, 45.
2. Cardillo, B., Gennaro, A., Merlini, L., Nasini, G. and Servi, S., *Phytochemistry*, 1973, 12, 2027.
3. Hayashi, S., Asakawa, Y., Ishida, T. and Matsuura, T., *Tet, Lett.*, 1967, 50, 5061.
4. Asakawa, Y., Hayashi, S. and Matsuura, T., *Experientia*, 1969, 25, 907.
5. Sofowora, E. A. and Hardmans, R., *Phytochemistry*, 1973, 12, 403.
6. Maheshwari, M. L. and Mukerjee, S. K., *Ibid.*, 1975, 14, 2083.

SYNTHESIS OF THERALIN

Nikonov *et al.*[1] have reported the isolation of 7, 4′-dihydroxy-2′-methoxy-isoflavone (theralin) in the grass and groats of *Thermopsis alterniflora*. Its structure is based on degradative and spectral studies. During the course of selective demethylation[2,3] and debenzylation studies at 2′-position of isoflavones, we have developed a convenient method for the synthesis of 2′-alkoxy isoflavones. The synthesis of theralin has been achieved through this procedure which confirms the above structure,



(I) $R = R_1 = R_2 = H$

(II) $R = R_1 = R_2 = CH_2C_6H_5$

(III) $R = R_1 = CH_2C_6H_5,\ R_2 = H$

(IV) $R = R_1 = CH_2C_6H_5,\ R_2 = CH_3$

(V) $R = R_1 = H,\ R_2 = CH_3$

Benzylation of 7, 2′, 4′-trihydroxy isoflavone[2] (I), m.p. 284° (d), with benzyl chloride gave the corresponding tribenzyloxy compound (II), m.p. 167° which on selective debenzylation at 2′-position using dioxan/HCl afforded 7, 4′-dibenzyloxy-2′-hydroxy isoflavone (III), in good yields, m.p. 175°. Methylation of (III) with dimethyl sulphate in acetone-K_2CO_3 yielded 7,4′-dibenzyloxy-2′,-methoxy isoflavone (IV), m.p. 133° which on debenzylation with pd/C yielded the required theralin (V) as light yellow needles, m.p. 276° (m.p. of natural theralin, 274–75°[1]). I.R. and U.V. spectra of the synthetic sample are in agreement with those reported for the natural sample.

Details of this work will the published elsewhere.

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---

1. Kattaev, N. S. and Nikonov, G. K., *Khim Prir Soedin*, 1975, 2, 140.
2. Aghoramurthy, K., Kukla, A. S. and Seshadri, T. R., *J. Indian Chem. Soc.*, 1961, 38, 914.
3. Kalra, V. K., Kukla, A. S. and Seshadri, T. R., *Indian J. Chem.*, 1967, 5 (7), 287.

PAPER CHROMATOGRAPHIC SEPARATION OF POTASSIUM, RUBIDIUM AND CAESIUM IN *n*-AMYL ALCOHOL-HYDROCHLORIC ACID-ETHANOL MIXTURE

THE solvent mixture (HCl-MtOH-BuOH-ketone) reported by Miller and Magee[1] for the paper chromatographic separation of K^+, Rb^+ and Cs^+ gives somewhat blurred bands. Phenolic solvents reported by Steel[2] and Magee and Headridge[3], though give sharp separations yet, attack the paper which becomes fragile and difficult to handle. Mixture of two of the straight chain aliphatic alcohols, or mixture of an alcohol and an acid has been successfully used for the paper chromatographic separation of Li^+, Na^+, K^+ and NH_4^+ ions[4-7]. Investigations were therefore undertaken for finding out the possibility of separation of K^+, Rb^+ and Cs^+ using a mixture of straight chain aliphatic alcohol and a mineral acid.

Experimental

Extra pure *n*-amyl alcohol, freshly distilled absolute alcohol and concentrated hydrochloric acid were used for the preparation of the solvent. Sodium lead cobaltous hexanitrate reagent[3] (fresh) was used as the spraying agent.

Development of chromatogram.— One end of 4 × 56·5 cm long strip of Whatman filter paper No. 4 was tailered to 2 cm width. On this tapered end, leaving about 8 cm margin, the requisite volume of the desired solution (0·1%) was applied as a narrow elongated spot (about 2·5 cm × 0·5 cm) and dried. The chromatograms were then developed by decending development till the solvent front moved to the other end of the paper strip. The developed chromatograms were dried in air, hanged upside down and sprayed first with ethanol and then with freshly prepared sodium lead-cobaltous hexanitrate reagent. After about five minutes, the excess of the reagent was washed out with ice-cold distilled water and the position of the bands (Brown for K^+ and Rb^+ and yellowish-brown for Cs^+) on the chromatogram was noted (Table I). R_f values of K^+, Rb^+ and Cs^+ in 1 : 2 : 1 AHE solvent determined at 20° ± 1° C are given in Table II.

Results and Discussion

In the paper chromatographic separation of K^+, Rb^+ and Cs^+, usually Cs^+ moves faster than the other two and therefore its separation from Rb^+ is comparatively easy. Hence the separation of K^+ and Rb^+ was first tried.

It will be seen from Table I that the best separation of K^+ and Rb^+ could be obtained by the solvent mixture No. 8, although No. 7 also gave clear separation of K^+ and Rb^+ bands with smaller band distance between the two. The R_f values of all the three alkali metal ions, *viz.*, K^+, Rb^+ and Cs^+ were therefore determined using solvent No. 8. It will be seen from Table II that the chromatogram of individual alkali metal gives higher R_f values than that of their values obtained from the chromatogram of the mixture of the three alkali metal ions in equal proportion.

Further studies on the separation of K^+, Rb Cs^+ using 1 : 2 : 1 :: AHE solvent mixture, on 4 cm

TABLE I

Effect of different solvent mixtures on the paper chromatographic separation of potassium and rubitium
(Load: 0·02 ml of solution containing 20 μg of each potassium and rubidium)

Sl. No.	Solvent mixture per cent by volume of			Time for development* hrs.	Position and width of		Space between bands	Remarks
	n-AmOH (A)	HCl (H)	EtOH (E)		K band cm	Rb band cm		
1.	..	40	60	20	7·0– 9·2	9·4–10·8	0·2	Partition of the bands observed
2.	60	40	..	27	8·0–12·0	..	Nil	
3.	..	50	50	16	13·0–15·5	16·0–18·2	0·5	Diffused bands; space between the bands not clear
4.	50	50	..	25	16·0–17·5	17·5–19·0	Nil	Partition of the bands observed
5.	30	50	20	18	16·5–18·0	18·5–21·0	0·5	Diffused bands; in between space not clear
6.	20	50	30	20	17·5–18·5	19·5–21·0	1·0	Bands slightly blurred
7.	20	40	40	25	9·5–11·0	12·0–13·5	1·0	Clear separation
8.	25	50	25	20	16·5–18·5	20·0–22·0	1·5	Clear separation
9.	30	40	30	26	8·4–10·7	11·2–13·2	0·5	
10.	40	40	20	26	7·8– 9·5	10·2–12·0	0·4	
11.	30	30	40	25	4·5– 5·5	6·0– 7·0	0·5	Diffused bands
12.	35	30	35	25	3·0– 5·0	..	Nil	No separation

* Time required to reach the solvent front upto the other end of the paper strip, *i.e.*, 48 cm.

TABLE II

R_f values of the K, Rb ana Cs and their separation in 1 : 2 : 1 AHE *solvent*

Expt. No.	Ion/ions loaded microgram	Width of the bands in cm			R_f values for		
		K	Rb	Cs	K	Rb	Cs
1.	30, K	18·3–20·7 (19·5)	..	..	0·410	..	..
2.	30, Rb	..	20·5–22·5 (21·5)	..	..	0·453	..
3.	30, Cs	..	..	24·5–26·0 (25·2)	..	..	0·531
4.	Mixture of K, Rb and Cs 50 μg of each	17·5–19·5 (18·5)	20·5–22·4 (21·5)	24·5–26·2 (25·4)	0·385	0·448	0·529
5.	do.	18·2–19·7 (18·9)	21·0–22·4 (21·7)	23·8–25·1 (24·5)	0·394	0·452	0·511

wide paper chromatogram, have revealed that (*i*) the minimum limits for the separation of K^+ and Rb^+ or Rb^+ and Cs^+ from their respective mixture is 20 μmg of any of the alkali from 500 μg of the others, and (*ii*) the maximum amount of the alkali metal ions that can be resolved under the said conditions is 600 μg mixture containing 200 μg of each of K^+, Rb^+ and Cs^+.

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Research Institute,
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May 27, 1976.

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---

1. Miller, C. C. and Magee, R. J., *J. Chem. Soc.*, 1951, p. 3183.
2. Steel, A. E., *Nature*, 1954, 173, 315.
3. Magee, R. J. and Headridge, J. B., *Analyst*, 1957, 82, 95.
4. Burstall, F. H., Davies, G. R., Linstead, R. P. and Wells, R. A., *J. Chem. Soc.*, 1950, p. 516.
5. Barnabas, T., Badve, M. G. and Barnabas, J., *Chem. Abstr.*, 1955, 49, 10793.
6. Sakaguchi, T. and Yasula, Y., *Ibid.*, 1952, 46, 3452.
7. Nakana, S., *Anal. Abstr.*, 1957, 4, 1100.

* * *

ON FISHER'S FUNDAMENTAL THEOREM OF NATURAL SELECTION WITH NON-OVERLAPPING GENERATIONS

THE Fundamental Theorem of Natural Selection first given by Fisher[1] broadly appears in two forms (Turner[2]). According to one form, the change in the average fitness of a population is equal to the genotypic variance in fitness. The other form, which includes the effect of a mating system, states that for random mating population, with two-allele system, the rate of increase in average fitness at any time is equal to its additive genetic variance at that time. Thus in the absence of dominance these two forms are identical. However, the interpretation of the dominance in a two-allele system is essentially that of an interaction between the two alleles, the three fitness values attached to the three genotypes forming an arithmetic series in the absence of dominance. Now it may happen that the three fitness values may form a geometric series so that on the logarithmic scale there would be no dominance although on the arithmetic scale some partial dominance will be exhibited resulting in two different forms of the theorem. The purpose of this communication is therefore to show that in situations where the dominance is due to scale effects, the two forms of the theorem would still be identical, although the variance due to dominance deviations on the arithmetic scale is not zero.

Let the relative fitness of the three genotypes *AA*, *Aa* and *aa* be respectively w_2, w_1 and w_0 in a random mating population with gene frequencies p for A and q for a with $p + q = 1$. The average fitness of such a population, denoted by $\bar{w}$ can be expressed as

$$\bar{w} = pw_2 + qw_0 - pq\,(w_2 - 2w_1 + w_0) \qquad (1)$$

where $(w_2 - 2\,w_1 + w_0)$ expresses the degree of dominance on the arithmetic scale. After the operation of natural selection the increase in average fitness denoted by $\triangle\,\bar{w}$ is

$$\triangle\,\bar{w} = \sigma_{\bar{w}}^2/\bar{w} \qquad (2)$$

where $\sigma_w{}^2$ is genotypic variance of the fitness values. Now the genotypic variance is the sum of additive and dominance variances which can be expressed respectively as

$$\sigma_A{}^2 = 2pq\,[\bar{w}\,(w_2 - 2w_1 + w_0) + (w_1{}^2 - w_2 w_0)] \qquad (3)$$

$$\sigma_D{}^2 = p^2q^2\,(w_2 - 2w_1 + w_0)^2. \qquad (4)$$

An alternative expression for the change in the average fitness can then be shown to be equal to

$$\triangle\,\bar{w} = \frac{\sigma_A{}^2}{\bar{w}}\left[1 + \frac{pq\,(w_2 - 2w_1 + w_0)}{2\,(\bar{w} + \theta)}\right] \qquad (5)$$

where

$$\partial = (w_1{}^2 - w_2\,w_0)/(w_2 - 2w_1 + w_0). \qquad (6)$$

Taking into account the round of random mating in addition to the effect of natural selection, the change in the average fitness of the population, denoted by $\triangle\,\bar{w}^*$ is, according to the derivations of Li[3] and using (1), given by

$$\triangle\,\bar{w}^* = \frac{\sigma_A{}^2}{2\bar{w}}\left[1 + \left(\frac{pw_2 + qw_0}{\bar{w}}\right)\right]$$

$$= \frac{\sigma_A{}^2}{\bar{w}}\left[1 + \frac{pq\,(w_2 - 2w_1 + w_0)}{2\bar{w}}\right]. \qquad (7)$$

Comparing the two expressions (5) and (7), it is clear that even if $(w_2 - 2\,w_1 + w_0)$ is not zero, the two $\triangle\,\bar{w}$ and $\triangle\,\bar{w}^*$ are identical provided $w_1{}^2 = w_2\,w_0$. This would mean that, on the geometric scale, there is no dominance.

It is apparent therefore that while the behaviour of the average fitness under natural selection without involving the effects of mating system is more rigorous, the change in average fitness, taking into account the random mating, is very much dependent on the scale on which the fitness values are measured. This also points out to the limitation of the concept of the additive genetic variance in that it involves the linearity assumption. On the other hand, the genotypic variance is a quantity free from any such assumption and therefore the Fundamental Theorem of Natural Selection employing it, *viz.*, in the form of (2) is much more general than its other form given by (7) which has been debated by several workers.

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Library Avenue,
New Delhi 110 012, *April* 23, 1976. PREM NARAIN.

---

1. Fisher, R. A., *The Genetical Theory of Natural Selection*, Clarendon Press, Oxford, 1930.
2. Turner, J. R. G., *Mathematical Topics in Population Genetics*, Edited by K. Kojima, Springer-Verlag, Berlin-Heidelberg, New York, 1970.
3. Li, C. C., *Nature*, London, 1967, 214, 505.

HISTOLYSIS OF PEDIPALPAL MUSCLE OF THE SCORPION *HETEROMETRUS FULVIPES* (C. KOCH) DURING MOLTING

LITERATURE on arachnid metabolism during molting is scarce. On the other hand ecdysial metabolism in insects and crustacea is extensively worked out. The present communication makes a preliminary report of hitolysis in the pedipalpal chelate muscle of the scorpion, *Heterometrus fulvipes*.

In our collection of the scorpions, we came across a female specimen, clearly in the post-molt condition. The animal was lethargic and the skin was soft and transparent. No attempt was made to stage the animal in the molt cycle as proposed by Drach[1]. The flexor-extensor muscle mass was exposed in the chela of the pedipalp by incision of the exoskeleton. It was granulated and had a more fluid appearance than that of the normal scorpion. This 'liquidized' appearance may be due to increased water content.

TABLE I

Pedipalpal chela muscle, weight, its histosomatic index and some biochemical parameters in relation to molting in the scorpion Heterometrus fulvipes

Parameter	Normal	Post-molt	*t*	d.f.	P	% Change
Muscle weight (mg/g wet)	[a]522 ±49·17 (4)	269 (1)	[b]4·602	3	<0·02	− 48·5
Histosomatic index of muscle (HSI)	82·18 ±3·416 (4)	3·461 (1)	1·245	3	NS	−57·9
Total protein (mg/g wet)	87·757±6·876 (4)	138·28 (1)	6·573	3	<0·01	+57·6
TFFPS (μg/g wet)	12·06 ±2·992 (4)	38·69 (1)	7·961	3	<0·005	+220·8
Total protein (Holohistontic levels, HHL) (mg/tissue)	45·60 ±2·771 (4)	37·20 (1)	2·711	3	NS	−18·4
TFFPS (HHL) (μg/tissue)	6·328±1·733 (4)	10·41 (1)	3·656	3	<0·05	+64·6

a. Values are means ± standard deviations (numbers of estimation).
b. Statistical treatment (according to Pillai and Sinha[11]).
t = Calculated students' *t* test values.
d.f. = Number of degrees of freedom.
P = Level of significance of difference. NS = Not significant.

A similar correlation between increased water content and granulation of muscle has been noted when the muscle atrophies due to deprivation of its innervation in the scorpion[2] and in the vertebrates[3-7]. It may, however, be noted that this comparison between liquidization of muscle during molting and that occurring due to denervation has to be confined only to the limited extent of physical appearance and increase in water content since the causo-mechanisms for these two processes are different: denervation atrophy involves an important deprivation of trophic influence of the nerve on the muscle whereas in liquidization associated with molting no such clear-cut trophic influences can be implicated.

The muscle total weight (holo-histontic weight) obtained by pooling the muscle masses of chelae of both pedipalpi of the scorpion, as also the histosomatic index (HSI, obtained by calculating the weight of chelate muscle per cent total body weight) show important decreases in the molted scorpion, although the decrease in the HSI is not statistically significant (Table I). The decrease of the above parameters are indications of high level of atrophy undergone by the muscle during molting. The total proteins, estimated with Folin-Ciocalteu reagent[8] in the residues of trichloroacetic acid (TCA) homogenates were significantly greater in the 'molted' muscle as compared to the normal muscle. However, the total protein content when expressed in terms of the whole tissue (holo-histontic level = total individual tissue level), showed decrease (− 18·4%). The total free Folin-positive substances

(TFFPS, which include free amino acids) estimated in TCA supernatants using Folin-Ciocalteu reagent in conjugation with cobalt reagent[9] showed increases in weight specific as well as holo-histontic levels in the 'molted' muscle. The granular and more fluid consistency of the molted muscle, the decrease of total protein content and increase of TFFPS (which include free amino acids) at the holo-histontic level are reasonable indications of atrophy of the muscle associated with molting.

The tissues predominantly concerned during crustacean ecdysis are blood, hepatopancreas and integument[10]. Nevertheless the muscle also undergoes certain changes. Extensive histolysis of the large chela muscle of *Gecarcinus lateralis* has been reported. This involves considerable loss of muscle protein[12]. The data presented here for *H. fulvipes* muscle involving decrease of total protein at holo-histontic level may be due to such histolytic process.

The degradation of the chela muscle tissue in *G. lateralis* has been suggested to aid in the removal of large muscle mass through narrow basal joint during ecdysis. It could be the same in scorpion.

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---

1. Drach, P., *Ann. Inst. Oceanog.* (*Paris N. S.*), 1939, 19, 391.
2. Satyanarayana, K., Selvarajan, V. R. and Swami, K. S., *Comp. Biochem. Physiol.*, 1975, 52, 386.
3. Oppenheim, H., *Handbuch. Heilkunde der Nerven*, Berlin, 1894.
4. Beattie, J. M. and Dickson, W. E. C., *A Text-book of Pathology*, London, 1943.
5. Adams, R. D., Denny Brown, D. and Pearson, C. M., *Diseases Muscle. A Study in Pathology*, 2nd Ed., P. B. Hoeber, N.Y., 1962.
6. Drahota, Z., "Electrolytes in denervated muscle," In *The Denervated Muscle* (Edited by Gutman, E.), Publishing House of the Ezechoslovak Academy of Sciences, Prague, 1962.
7. Humolier, H. L., Griswold, P. and McIntyre, A. R., *Ann. J. Physiol.*, 1950, **161**, 496.
8. Lowry, O. H., Rosebrough, N. J., Farr, A. L. and Randall, R. J., *J. Biol. Chem.*, 1951, 193, 275.
9. Matsushita Setsuro, Nobuko Iwami and Yuki Nitta, *Anal. Biochem.*, 1966, 16, 371.
10. Yamaoka, L. H. and Scheer, B. T., In *Chemical Zoology*, Vol. VI, Chapter 11, Ed. by Florkin and Scheer, A. P., N.Y., 1971.
11. Pillai, S. K. and Sinha, H. C., *Statistical Methods for Biological Workers*, Ram Prasad & Sons, Agra-3, 1968.
12. Skinner, D. M., *J. Exptl. Zool.*, 1966, **163**, 124.

A COMPARISON OF THE EFFECT OF CYCLOHEXIMIDE TREATMENT ON THE ANTIVIRAL ACTIVITY OF DIFFERENT TYPES OF INTERFERON INDUCERS IN MICE

INTERFERON induction and antiviral activity of Newcastle disease virus (NDV)[1,2] have already been reported. Tests with 6–MFA, an antiviral agent from *Aspergillus flavus*. DU/KR/162 *b* (now re-identified by Dr. D. I. Fennell, NRRL, Peoria, Illinois, U.S.A. as *Aspergillus ochraceus*, Personal communication) showed that mice treated with 6–MFA produce serum and tissue interferon (unpublished data) and also develop anti-Semliki Forest virus resistance[3-5]. A subculture of the fungus has been deposited with the American Type Culture Collection (ATCC 28706).

Evidence over the past few years has accumulated to show that interferon production stimulated by viral or nonviral inducers can significantly be enhanced by cycloheximide treatment in animals[6] as well as in cultured tissue cells[7-8]. Because of a possible clinical significance of this, we have compared the inducing capacity of antiviral resistance of 6–MFA in mice with that of NDV in the presence of cycloheximide and have noticed important differences.

Thirty-five day old Swiss-CDRI mice of either sex weighing 16–18 g were used in the present experiments. Semliki Forest Viruh (SFV) was procured from the ATCC and passaged intracerebrally (i.c.) in the 35 day old mice. A 10% brain homogenate was prepared in Hanks' balanced salt solution, which formed the stock pool and maintained in aliquots at – 20° C. The strain of neurovaccinia virus was procured from the ATCC and maintained similarly. The LD_{50} titres of both viruses were calculated by the Reed and Muench method[9], on the basis of ten-fold dilutions of inocula.

The egg-adapted vaccine (allantoic fluid) strain of NDV was obtained from the Animal Husbandry Department, Lucknow. 6–MFA was prepared by acetone treatment of the crude filtrate of *A. ochraceus* as described earlier[3]. Cycloheximide was

TABLE I

Proportion of mice surviving vaccinia/SFV challenge, following injection with two inducers (6–MFA or NDV) in presence of cycloheximide

Treatment		Interferon Inducers							
		NDV (410 HA units/0·2 ml)				6–MFA			
		IHD CHALLENGE		SFV CHALLENGE		IHD CHALLENGE		SFV CHALLENGE	
		Survivors*	Survival (%)	Survivors	Survival (%)	Survivors	Survival (%)	Survivors	Survival (%)
Cycloheximide** 1 *hr before* inducer		0/10	0	9/18	50	2/8	2	1/10	10
Cycloheximide *along with* inducer		3/9	33	0/18	0	0/10	0	2/9	22
Cycloheximide 1 *hr after* inducer		0/10	0	6/18	33	1/10	10	4/10	40
Only Cycloheximide)	Controls	0/10	0	2/10	20	0/10	0	0/10	0
Only Inducer		0/10	0	5/10	5/10	2/8	25****	0/10***	0
Only PBS+		0/10	0	5/10	50	0/10	0	0/10	0

* Mice surviving/total number.
** Cycloheximide injected at the rate of 1 mg/mouse.
*** 6–MFA injected at the rate of 0·1 mg/mouse.
**** 6–MFA injected at the rate of 1·0 mg/mouse.
+ Phosphate buffer saline.

purchased from the Upjohn Company, Kalamazoo, Michigan, U.S.A. It was given at the rate of 1 mg/mouse i.p.

Mice were injected with sub-protective doses of 6–MFA or NDV (R$_2$B strain) by the intraperitoneal (i.p.) route. Cycloheximide was given 1 h. *before*, simultaneously, or 1 h. *after* the inducer. Twenty-four hours following the administration of the inducer, all the animals were challenged with SFV, subcutaneously (s.c.), or with IHD vaccinia virus by the i.c. route. Animals were observed for 14 days for specific symptoms of encephalitis and death. The percentage of mice surviving fatal virus challenge was calculated in terms of the control.

Results presented in Table I showed that with NDV (inducer) given to mice at a subeffective dose (410 HA units/0·2 ml) that confers *no* protection to challenge fatal neurovaccinia virus (100 LD$_{50}$, 0·03 ml, i.c.), greater protection (33%) of the animals was obtained when cycloheximide was given *along with* the inducer. However, with 6–MFA as the inducer, a *depressive effect* (lowered protection) of cycloheximide was noticed : 6–MFA, at a dose of 1 mg/mouse, protected only 25% of the animals challenged with a lethal dose (100 LD$_{50}$, 0·03 ml, i.c.) of IHD virus ; the protection rate fell to zero, if cycloheximide was administered along with 6–MFA.

NDV given at a rate of 410 HA units/0·2 ml per mouse did not protect any animal challenged with a low dose (1 LD$_{50}$, 0·5 ml s.c.) of SFV which itself caused only 50% mortality in the untreated control. If cycloheximide was given *along with* NDV, the protection rate fell to zero. However, with 6–MFA as the inducer, injection of cycloheximide *enhanced* the antiviral activity (10 to 40%) of a dose of 6–MFA (0·1 mg/mouse), which only increased the mean survival time of the mice against a lethal challenge with SFV (100 LD$_{50}$, 0·5 ml, s.c.).

Thus, the effect of cycloheximide in increasing or decreasing the antiviral activity of interferon-inducing agents is here seen to vary not only with the inducer used, but also with the challenge virus employed. The time of administration of cycloheximide also plays an important role in connection with the protection level.

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1. Henle, W., Henle, G., Denihardt, F. and Berges, V. V., *J. Exp. Med.*, 1959, **110**, 525.
2. Youngner, J. S., Scott, A. W., Hallum. J. V. and Stinebring, W. R., *J. Bacteriol.*, 1966, **92** (4), 862.
3. Maheshwari, R. K. and Gupta, B. M., *J. Antibiotics*, 1973 *a*, **26** (6), 320.
4. — and —, *Ibid.*, 1973 *b*, **26** (6), 328.
5. — and —, *Ibid.*, 1973 *c*, **26** (6), 335.
6. DeClerq, E. and Merigan, T. C., *Virology*, 1970, **42**, 799.
7. Tan, Y. H., Armstrong, J. A., Ke, Y. H. and Ho, M., *Proc. U.S. Nat. Acad. Sci.*, 1970, **67**, 464.
8. Ho, M., Tan, Y. H. and Armsrong, J. A., *Proc. Soc. Exp. Biol. Med.*, 1972, **139**, 259.
9. Reed, L. J. and Muench, H., *Am. J. Hyg.*, 1938, **27**, 493.

SUSCEPTIBILITY OF MOUSE PERITONEAL MACROPHAGES TO INFECTION WITH ARBOVIRUSES

MACROPHAGE cultures from normal mice support the growth of mouse hepatitis viruses[2-4] and peritoneal macrophages (PM) from ectromelia-immune[1] mice support the growth of ectromelia viruses. Two arboviruses, *viz.*, West Nile (WN) and Yellow Fever (YF) have been shown to grow in mouse PM[5]. We report the results of susceptibility studies of mouse PM to thirteen different arboviruses isolated in India.

In the preliminary experiments, two ml of 2% starch[5] (in normal saline) was injected into mice by the intraperitoneal (IP) route (on the previous day) to activate the macrophages. In further experiments no such activator was used, because the yield of PM was adequate without its use. Three ml of sterile Hanks' balanced salt solution[6] was injected IP into 8–10 week old male Swiss albino mice and after kneading the abdomen gently, the IP fluid from each mouse was collected in a culture tube. After two hours of incubation at 37° C, saline was removed and the tubes fed with medium M 199 (Earle's), supplemented with 20% goat serum, loosely stoppered and incubated at 37° C in 5% CO$_2$ atmosphere[6]. After 24 hours, tubes with well-spread macrophages were selected and washed with the medium, tightly stoppered and incubated at 37° C for 48 hours. For the experiment, twenty tubes were selected and taken for inoculation of one virus. The viruses employed (Table I) were diluted in M 199 to contain approximately 3–4 dex[7] of infant mouse LD$_{50}$ or TCID$_{50}$ virus and the tubes were inoculated with 0·1 ml of virus suspension per tube.

TABLE I

Antigenic group	Name of the virus, abbreviation strain number and passage history
Group A	Sindbis (SIN) (AR 339) M_5V_2; Chikungunya(CHIK) (634029) $M_{11}V_1$
Group B	Japanese encephalitis (JE) (P 20778) M_9; Kyasanur Forest disease virus (KFD) (P 9605) M_{15}; West Nile (WN)* (E 101) M_5V_2
VSV	Chandipura (CHP) (653514) $M_{22}V_1$
Kaisodi	Kaisodi (KSO) (G 14132) M_{43}
Bunyamwera	Batai (BAT) (G 20217) $M_{12}V_1$
Nairobi Sheep Disease	Ganjam (GAN)+ (G 619) M_5BH_1
Simbu	Ingwavuma (ING) (633970) M_7BH_1; Sathuperi (SAT) (G 11155) M_{13}
Ungrouped	Wanowrie (WAN) (G 700) M_{19}; Bhanja (BHA) (G 690) M_5BH_1

* Not isolated at Virus Research Centre.
+Renamed Nairobi Sheep Disease (NSD) since 1975.
M = Suckling mouse brain passage.
V = Vero passage.
BH = Baby hamster kidney (BHK-21) passage.

After adsorbing the virus for one hour at 37° C, the tubes were washed five times with neutralized Hanks' BSS and fed with M 199 with 10% goat serum and incubated at 37° C.

The tubes were observed daily for cytopathic effect (CPE) upto ten days. On the zero, third, seventh and tenth days of post-inoculation (PI) about 4–5 tubes were taken out and stored at – 50° C for a maximum period of one week. The tubes were subjected to three cycles of freezing and thawing and centrifuged at 250 g for 10 mins. The supernatant fluid was assayed in 2–3 day old infant mice by the intracerebral (IC) route. The sick mice were harvested and 10% brain suspension prepared in normal saline was tested by the quick complement fixation test[8] for identification of the viruses.

Out of the thirteen arboviruses tested, only Ingwavuma (ING) (633970) virus showed evidence of CPE and multiplication. The CPE was in the form of rounding of 75% cells on the second PI day and 100% on the next. Although the titre of the ING virus was not very high, the second passage tissue culture fluid, when titrated in infant mice, showed evidence of multiplication. The WN virus multiplied without any CPE; Kyasanur Forest Disease (KFD) and CHP viruses persisted in traces upto the 10th PI day, but could not be detected on subsequent passage. The remaining viruses did not show any evidence of multiplication, when the infected tissue culture fluids of the zero, 3rd, 7th and 10th PI days were assayed in infant mice.

It appeared that mouse PM supported the growth of only WN and ING viruses from among the arboviruses tested. This was further confirmed by carrying out three serial passages of these viruses in PM.

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---

1. Roberts, J. A., *J. Immunol.*, 1964, 92, 837.
2. Bang, F. B. and Warwick, A., *Virology*, 1959, 9, 715.
3. Mallucci, L., *Ibid.*, 1965, 25, 30.
4. Shif, I. and Bang, F. B., *J. Expt. Med.*, 1970, 131, 843.
5. Seamer, J., *Arch. ges. Virusforsch.*, 1965, 17, 654.
6. Chang, Y. T., *J. Nat. Cancer. Inst.*, 1964, 32, 19.
7. Haldane, J. B. S., *Nature*, 1960, 187, 879.
8. Pavri, K. M. and Sheikh. B. H., *Curr. Sci.*, 1966, 35, 455.

ANTIVIRAL ACTIVITY OF 6MFA, GROWTH PRODUCT OF FUNGUS *A. OCHRACEUS*, AGAINST FOOT AND MOUTH DISEASE VIRUS

Foot and Mouth Disease (FMD) is an acute viral disease of the cloven footed animals. The virus comes in seven major types, namely, 'O', 'A', 'C', 'Asia-1', 'SAT-1', 'SAT-2' and 'SAT-3 and has over 62 subtypes.

Use of interferon (IF) and interferon inducers to control and manage FMD (picornavirus) virus infection in animals has been considered seriously in recent years. Particular mention may be made of yeast *RNA*, a product which, if injected continuously into infected mice, guinea pigs or cattle, is claimed to cause delayed clinical manifestations of FMD[1] presumably caused by interferon. Because virulent strains of FMD seem ot produce less of IF in host animals[2] and the avirulent strains more of it, there is a suggestion that FMD virus multiplication in mice can be restricted if adequate concentrations of IF in serum or body tissues is maintained through endogenous production or exogenous augmentation.

We recently reported isolation of an inducer of interferon trivially designated as 6–MFA[3-5] obtained from the fungus *Aspergillus flavus* Du/KR/162 *b* (Syn. *A. ochraceus* ATCC 28706), which inhibits Semliki Forest Virus (SFV) infections in experimentally inoculated Swiss Albino mice. We now report the results of tests of antiviral activity of 6–MFA against FMD virus.

6–MFA, a polysaccharide nucleoprotein complex, was prepared as per the technique described by Maheshwari and Gupta[3]. FMD virus 'O' and 'A' were obtained from IVRI in the form of 10% (w/v) muscle homogenates of the infected mice. Both strains are well adapted to multiply in 2–3 weeks old mouse[6]. For 'O' strain the MST values were 4·8 and 5·1 days, and for 'A' they were 4·5 and 4·5 days (*see* Table I). To assess anti-FMD virus activity, mice were first treated with 6–MFA through intraperitoneal route at the rate of 2 ml and 1 ml doses per mouse. This quantity of 6–MFA, chosen to treat mice against FMD, corresponds to that which usually gives 80% protection against SFV infection in similar mice[3]. Treated animals were challenged 24 hrs. later with the viruses ('O' and 'A') given by intramuscular route at the rate of 0·25 ml of $10^{2\cdot6}$ and $10^{1\cdot6}$ of type 'O', and $10^{2\cdot2}$ and $10^{1\cdot2}$ of type 'A' respectively (100 and 1000 LD$_{50}$) per mouse. The LD$_{50}$ contained in the inoculum was determined by Reed and Muench[7] method. Mice were observed twice daily morning and evening for the development of specific paralytic symptoms and death upto 14 days after which the experiments were terminated.

In general, the proportion of animals protected against type 'O' was more than in type 'A' indicating that FMD type 'O' virus appears to be *more sensitive* to the antiviral action of 6–MFA than the type 'A'. Even then, the MST of treated mice (type 'A' infected animals) has been found to vary directly with the quantity of the inducer (Table I). Furthermore, the MST here was seen to increase with the size (100 and 1000 LD$_{50}$) of the challenge inoculum. This type of response can be explained on the basis of the participation of a specific type of immune (antibody) response increasing with

TABLE I

Showing the antiviral activity of 6-MFA against the FMD virus %

FMD virus type	Dose of 6MFA/PBS (i.p.)	No. of mice	Virus (i.m.)	Protection S/T (%)	MST (in days)
'O'	1 ml	9	100 LD_{50}	3/9 (33·3)	6·5
	2 ml	9	,,	5/9 (55·5)	7·5
	1 ml PBS (CONTROL)	10	,,	Nil	4·8
	1 ml	10	1000 LD_{50}	4/10 (40·0)	7·3
	2 ml	10	,,	3/10 (30·0)	6·5
	2 ml PBS (CONTROL)	10	,,	Nil	5·1
'A'	1 ml	9	100 LD_5	1 9 (11·1)	5·0
	2 ml	9	,,	1/9 (11·1)	6·6
	1 ml PBS (CONTROL)	10	,,	Nil	4·5
	1 ml	10	1000 LD_{60}	1/10 (10·0)	7·5
	2 ml	8	,,	3/8 (37·5)	8·0
	2 ml PBS (CONTROL)	10	,,	Nil	4·5

S = number of surviving; T = total mice. i.p. = intraperitoneal; i.m. = intramuscular.
PBS = phosphate buffer saline.

increased supply of the antigen apparently accumulating through intracellular antiviral action of IF induced by 6–MFA.

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1. Thely, M., Choay, J., Dhenin (L) ouis, and Dhenin (L) e'one., *Compt Rend.*, 1963, 256, 1048 ; cited by Bachrach, H. L., *Ann. Rev. Microbiol.*, 1968, 22, 201.
2. Zygraich, N. and Williams, R., *Bull. Off Int. Epizoot.*, 1967, 67, 731 ; cited by Bachrach, H. L., *Ibid.*, 1968.
3. Maheshwari, R. K. and Gupta, B. M., *J. Antibiotics*, 1973 *a*, 26 (6), 320.
4. — and —, *Ibid.*, 1973 *b*, 26 (6), 328.
5. — and —, *Ibid.*, 1973 *c*, 26 (6) 335.
6. Rao, B. U., Thesis submitted for the degree of Ph.D. in Bacteriology and Virology in Agra University, 1973.
7. Reed, L. J. and Muench, H., *Am. J. Hyg.*, 1938, 27, 493.

STREPTOMYCES VIJAENSIS—A NEW STREPTOMYCETE

WHILE carrying out the systematic screening for the isolation of streptomycetes capable of producing new antibiotics, a new streptomycete was isolated from a soil sample collected from Krishna river bed at Vijayawada (A.P.) which was designated as *S. vijaensis*.

The materials and methods used were similar to those of our previous communication[1] and ISP procedures[2]. The sporophores are simple, and occurred as straight to wavy in most cases with a few extended open spirals and occasionally a few open and closed hooks. No verticils were observed. The spores are oval to elliptical with warty surface.

Cultural characters.—Glucose-Czapek's agar : Growth excellent ; reverse yellowish brown to brown (14–A–10) ; aerial mycelium abundant, pale dull rose (3–A–8) ; yellowish brown throughout and purple (43–F–7) pigment around colonies.

Glycerol-asparagine agar ISP : Growth good ; reverse colourless to light yellowish brown (12–G–6) ; aerial mycelium moderate, pale pink (2–B–1).

Inorganic salts-starch agar ISP : Growth excellent ; reverse light dull brownish yellow (11–C–3) ; aerial mycelium abundant, light dull rose (4–A–8).

Oatmeal agar ISP : Growth good ; reserve cream ; aerial mycelium good, pale pink (2–C–1).

Potato plug: Growth excellent, wrinkled; mycelium pale reddish brown; aerial mycelium poor, white with pale rose patches; plug turned to deep blackish brown.

Nutrien agar: Growth good; reverse yellowish brown to brown (14–F–9); aerial mycelium scant white; deep brown soluble pigment.

Yeast extract-malt extract agar ISP: Growth good; reverse brown (12–H–10); aerial mycelium good, light dull pink (4–D–1); light brown soluble pigment.

The following biochemical reactions were positive: melanin, H_2S, tyrosinase, amylolytic activity, nitrate reduction, milk coagulation with acidic reaction, acid from glucose, catalase formation, gelatin liquefaction, casein hydrolysis, cellulolytic activity (slight) and sucrose inversion. The following reactions were negative: Indole production from tryptone, Loeffler's serum liquefaction, blood haemolysis and milk peptonization.

Carbon utilisation: *Excellent to good growth*: arabinose, fructose, glucose, galactose, inulin, inositol, maltose, mannitol, mannose, lactose, rhamnose, starch, sorbitol, sucrose, salicin and xylose. *Moderate to poor growth*: cellulose, glycerol, raffinose, sodium acetate, sodium citrate and sodium succinate. *Doubtful growth*: Dulcitol, digitonin and sorbose.

No growth: paraffin. Although the isolate broadly possesses poor antibiotic activity, the extracellular metabolite showed good activity on *P. vulgaris* and *S. typhosa* and the intracellular metabolite showed good activity on coagulase positive *Staph. aureus*.

Discussion

A detailed survey of the literature indicated that our isolate is related to *S. griscoroseus*[3] and *S. roseochromogenes*[4,5] and authentic cultures of these two were obtained and comparative studies made. The data show that *S. vijaensis* more closely resembles *S. griseoroseus* than *S. roscochromogenes*. However, even *S. griseoroseus*, although similar, differs from our isolate in (1) tyrosinase negative, (2) positive peptonization of milk and (3) no utilisation of raffinose, sorbitol, inositol and inulin.

In view of the above, our isolate is considered a new species and designated *S. vijaensis* sp. nov., as its origin in Vijayawada.

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---

1. Sambamurthy, K. and Ellaiah, P., *Hindustan Antibiot. Bull.*, 1974, 17, 24.
2. Shirling, E. B. and Gottlieb, D., *Int. J. Syst. Bacteriol.*, 1966, 16, 313.
3. Heinemann, B., Gourevitch, A., Lein, J., Johnson, D. L., Kaplan, M. A., Vanas, D. and Hooper, I. R., *Antibiot. Ann.*, 1954–55, p. 728.
4. Waksman, S. A., *The Actinomycetes*, Vol. 2, *Classification, Identification and Descriptions of Genera and Species*, The Williams and Wilkins. Co., Baltimore, 1961.
5. Shirling, E. B. and Gottlieb, D., *Int. J. Syst. Bacteriol.*, 1968, 18, 69; 1968, 18, 278; 1969, 19, 319.

NITROGEN FIXATION BY *AZOTOBACTER* OCCURRING IN THE RHIZOSPHERE OF C_4 PLANT SPECIES

THE non-symbiotic nitrogen fixing bacterium, *Azotobacter*, is receiving greater recognition among scientists[1,2]. The rank and luxuriant growth of certain tropical grasses like *Paspalum notatum*, *Digitaria decumbens* Hall., *Cynodon dactylon* Pers. and *Zea mays* L. is believed to be due to the association of active and efficient *Azotobacter* sp. in their root system[3]. Such plant species strangely enough exhibit an alternative but more efficient mechanism of carbon dioxide fixation called "C_4 pathway" (C_4-dicarboxylic acid pathway)[4] quite distinct from the universally functioning Calvin's pathway. In the present communication the N-fixing efficiency of several isolates of *Azotobecter* obtained from C_3 and C_4 plant species is compared.

From the rhizosphere of three species of C_4 plants (Table I) and from three C_3 plant species (Table II) *Azotobacter* cultures were isolated following standard microbiological techniques[5] and they were maintained on Waksman No. 77 agar medium. The N-fixing ability of the isolates was determined following the procedures detailed by Rangaswami and Subba Raja[6]. The total nitrogen in the broth after 10 days of growth of the organism under shake culture conditions was determined in a micro-Kjeldhal unit[7]. The respiratory activity of the isolates was measured in a Warburg respirometer[8]. The growth-promoting efficiency of *Azotobacter* isolates was tested on three rice varieties following the paper roll towel seed germination technique[9]. The results are presented in Tables I to III.

TABLE I

Nitrogen fixing efficiency of Azotobacter *isolates*

Source of *Azotobacter*	Isolate No.	mg of nitrogen fixed/g of mannite
C_3 *Plants*:		
Oryza sativa L.	AB. 1	16·80
	AB. 2	18·00
	AB. 3	18·60
	AB. 4	11·50
	AB. 5	12·30
Gossypium arboreum L.	GA. 1	7·64
Eleusine coracana Gaertn.	EC. 1	8·93
	EC. 2	8·33
Capsicum annuum L.	CA. 1	9·52
C_4 *Plants*:		
Amaranthus viridis L.	Am. 1	19·60
	Am. 2	22·40
	Am. 3	19·80
Brachiaria mutica Stapf.	Bm. 1	12·50
	Bm. 2	22·40
Cyperus rotundus L.	Cyp. 1	22·40
	Cyp. 2	14·00
	Cyp. 3	25·20
	Cyp. 4	19·60

TABLE II

Respiratory activity of Azotobacter

Isolate No.	Respiratory activity* (μ moles of O_2 consumed/g of cell/hr)
AB. 2	249
AB. 3	68
Am. 1	260
Am. 2	229
Am. 3	102
Bm. 1	246
Bm. 2	110
Cyp. 1	139
Cyp. 2	186
Cyp. 3	622
Cyp. 4	485

* Data represent average of two estimations.

TABLE III

Effect of seed inoculation with Azotobacter *on three rice varieties*

Source of *Azotobacter*	Vaigai		IR. 20		Bhavani	
	Plumule (mm)	Radicle (mm)	Plumule (mm)	Radicle (mm)	Plumule (mm)	Radicle (mm)
1. C_3 Isolates*	16·2	42·3	6·5	25·7	12·3	30·5
2. C_4 Isolates*	26·1	51·7	7·0	30·0	14·8	34·3
3. Water control	15·6	40·6	4·0	19·7	8·6	27·7

Data represent average of 20 seedlings measured five days after germination.
* Average values for nine isolates.

The data clearly reveal that many of the *Azotobacter* isolates from the C_4 plants have proved to be very efficient in N-fixation as compared to the isolates from the C_3 plants like rice, cotton, chillies and ragi. The isolate from *Cyperus rotundus* L. has fixed the maximum quantity of nitrogen and the minimum was by GA. 1 from cotton. Admittedly, a few *Azotobacter* isolates like AB. 1, AB. 2 and AB. 3 have exhibited greater N-fixing ability than the isolates, Bm. 1 and Cyp. 2, obtained from C_4 plants. Among the isolates, Cyp. 4 has recorded the maximum respiratory activity and the least by AB. 3. The isolate, AB. 2, though from a C_3 plant species, exhibited a high respiratory activity comparable to that of C_4 isolates. It is quite reasonable that as suggested by Hill *et al.*[10], the "oxygenlabile nitrogenase" is conferred protection through the exceedingly higher respiratory activity.

The data on the response of three rice varieties to seed inoculation with *Azotobacter* are revealing. The isolates from the C_4 plants in general promoted the growth of rice seedlings much more than the isolates from the C_3 plant species. This observation is amply supported by the recent work of Iswaran and Sen[11] who inoculated the rice seedlings of the variety, Jaya with a culture of *Azotobacter* isolated from the roots of *Cyperus rotundus* L. and obtained increased grain yields. The increased growth of rice seedlings observed in the present study may be due to the cumulative effects of both N-fixation and the synthesis of growth-promoting substances[12] and this area of investigation is receiving our attention. It may therefore be generalised that the *Azotobacter* isolates from the C_4 plant species are not only more efficient, but also exhibit more growth-promoting effect than the isolates from the C_3 plants.

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1. Rubenchik, L. I., *Azotobacter and its Use in Agriculture*, Natl. Sci. Found., Washington D. C. and Israel Program for Scientific Translation Ltd., 1963, p. 278.
2. Postgate, J. R., *The Chemistry and Biochemistry of Nitrogen Fixation,* (Ed.), J. R. Postgate, Plenum Press, London, New York, 1971, p. 161.
3. Mulder, E. G. and Brotonegoro, S., In : A. Quispel (Ed.), *The Biology of Nitrogen Fixation,* North Holland Pub. Co., Amsterdam, Oxford, 1974, p. 37.
4. Hatch, M. D. and Slack, C. R., *Biochem. J.*, 1966, 101, 13.
5. Pramer, D. and Schmidt, E. L., *Experimental Soil Microbiology,* Burgess Pub. Co., Minneapolis, Minn., 1966, p. 107.
6. Rangaswami, G. and Subba Raja, K. T., *Proc. Indian Acad. Sci.*, 1962, 56, 174.
7. Bremner, J. M., *J. Agr. Res. Sci.*, 1960, 55, 11.
8. Umbreit, W. W., Burris, R. P. and Stauffer, J. F., *Manometric Techniques,* Burgess Pub. Co., Minneapolis, Minn., 1964, p. 338.
9. Anonymous, *Proc. Int. Seed Test Assoc.*, 1966, 31, 152.
10. Hill, S., Drozd, J. W. and Postgate, J. R., *Appl. Chem. Biotech.*, 1972, 22, 541.
11. Iswaran, V. and Sen, A., *Food Fmg. Agri.*, 1976, 7, 17.
12. Burlingham, S. K., *Ann. Report, Rothamstead Expt. Sta.*, 1964, p. 92.

INHIBITION OF INDUCTION OF STREPTOZOTOCIN DIABETES BY MANNOHEPTULOSE

Streptozotocin (SZN), derived from *Streptomyces achromogenes*, induces diabetes by destruction of the beta cells of pancreatic islets[12]. Chemically, it is 2-deoxy, -2- (3-methyl-nitrosoureido) -D- glucopyranose[9]. Its diabetogenic action is prevented by 2-deoxyglucose (2DG)[6] which is an inhibitor of insulin release[10] and of glucose metabolism[13]. This, and some other observations[2,5], suggest that the betacytotoxic action of SZN is related to insulin release or glucose metabolism. Against this possibility is an observation[6] that SZN diabetes was not prevented by mannoheptulose (MH), a more potent inhibitor of insulin release[3] and glucose metabolism[4]. In view of the theoretical importance of this negative finding, we have investigated again the interaction between MH and SZN.

The diabetogenic action of a single injection of SZN (gift from Upjohn Co.) at different time intervals after a dose of MH (from Sigma Chemicals Co.) was determined, comparing the glucose tolerance of the treated male albino mice (20 to 25 g) with that of untreated controls and of mice receiving the same dose of SZN alone. Glucose tolerance was estimated as the area (GTA) under the glucose tolerance curve, after a glucose load of 3 g/kg after 16 hours of fasting. Blood samples collected from the orbital sinus at 0, 60 and 120 min after glucose injection were analysed for reducing sugar[1]. The mean control GTA was 228 mg hr/100 ml. (± 19, S.D., in 24 mice). The increase (*i.e.*, Δ GTA) over this value, on the fifth day after treatment was a measure of the resultant diabetes and was calculated as percentage (% Δ GTA) of the mean control GTA. SZN (in citrate buffer at 5° C, pH 4·5), MH (in isotonic saline at 5° C) and glucose (in water) were given intraperitoneally in volumes of 0·01 ml./g body weight.

Mice which received SZN alone (Group A, Table I) or along with MH (Group B) became equally diabetic. But when MH preceded SZN with increasing time intervals (Group C, D, E), there was progressive reduction in the effect of SZN. With an interval of 60 min (Group E), diabetes was completely prevented. At 75 min (Group F), MH became altogether ineffective. Thus the protective action of MH against SZN was very much time dependent and the optimal time for MH administration was 60 min before SZN. A slightly lower dose of MH (125 mg/kg, Group G) was ineffective against the same dose of SZN (125 mg/kg). Thus the minimal dose of MH effective against 125 mg/kg SZN was 150 mg/kg. This dose was equally effective against a more severely diabetogenic dose of SZN (175 mg/kg, Groups H and I).

The present study establishes that MH can prevent the diabetogenic action of SZN. The preferential uptake of SZN by mouse beta cells is related to the glucose moiety in this nitrosourea[11]. The entry of MH into mouse beta cells is also by a glucose sensitive transport mechanism[8]. But the protective action of MH against SZN is not by competition for a common transport mechanism, because the protection is offered not when the circulating MH : SZN ratio would have been the highest (Group B), but 60 min later (Group E), by which time the

TABLE I

Effect of MH on the Diabetogenic action of SZN

Group* No.	Dose of SZN mg/kg	Mannoheptulose		GTA±SD mg hr/ 100 ml.	% △ GTA
		Dose, mg/kg	Time before SZN, min		
A	125	0	..	640±17	+180
B	125	150	0	642±16	+181
C	125	150	30	587±22	+157
D	125	150	45	492±19	+115
E	125	150	60	229±21	0
F	125	150	75	619±17	+171
G	125	125	60	649±10	+184
H	175	0	..	930± 9	+407
I	175	150	60	230±20	0

* All groups consisted of 6 mice each. For abbreviations and experimental details, *see* text.

blood concentration of MH would have fallen by half[3]. 60 min after intravenous administration of MH, maximal hyperglycaemia developed in rabbits[3], as also almost complete disappearance of circulating insulin in rats[7]. These considerations suggest a non-competitive mechanism for the action of MH. The similarity between MH and 2DG in preventing SZN diabetes, strengthens the possibility that the mechanism of action of SZN might be related to either glucose metabolism or insulin release. The failure of MH to protect rats against SZN in a previous study[6] is best explained by the very short time interval (15 min) by which the sugar had preceded SZN. The apparent constancy of the minimal effective dose of MH against varying doses of SZN is also interesting and is being investigated.

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1. Astoor, A. M. and King, E. J., *Biochem. J.*, 1954, **56**, 44.
2. Brosky, G. and Logothetopoulos, J., *Diabetes*, 1969, **18**, 606.
3. Coore, H. G. and Randle, P. J., *Nature*, 1963, **197**, 1264.
4. —, and —, 1964, **91**, 56.
and Burr, L. N., Diabetes, 1974, **23**, 232.
5. Culbert, S., Sharp, R., Rogers, M., Felts, P.
6. Dulin, W. E. and Beatrice, M. W., *Ibid.*, 1969, **18**, 459.
7. Foresch, E. R., Jacob, A., Zahind, G. R. and Simon, E., *Diabetologia*, 1966, **2**, 265.
8. Hellman, B., Idhal, L. A., Lernmark, A., Schlin, J., Simon, E. and Taljedal, I. B., *Molecular Pharmacology*, 1972, **8**, 1.
9. Herr, R. R., Jahnke, H. R. and Argoudelis, A. D., *J. Amer. Chem. Soc.*, 1967, **89**, 4808.
10. Kilo, C., Long, C. L., Bailey, R. and Recant, L., *Diabetes*, 1967, **16**, 377.
11. Hinz, M. and Pfeiffer, E. F., *Ibid*, 1974, **23**, Supplement I.
12. Rakietan, N., Rakietan, N. L. and Nadkarni, M. V., *Cancer Chemother. Rev.*, 1963, **29**, 91.
13. Wick, A. W., Drury, D. R., Nakada, H. I. and Wolfe, J. B., *J. Biol. Chem.*, 1957, **224**, 963.

SHORT SCIENTIFIC NOTES

A Note on the Occurrence of *Cercospora bidentis* Tharp. on *Helianthus tuberosus* L. in India

In November, 1975 plants of *Helianthus tuberosus* were found to be affected by a foliar disease. The disease was characterized by minute leaf-spots, up to 5 mm in diameter, irregular, amphigenous, brown with a light yellow chlorotic periphery, coalesce to form blighted patches mostly along the margin and at the tip of the leaf. Mycelium immersed, hyphae colourless forming pseudostroma in the sub-stomatal cavity. Fruiting mostly epiphyllous, stroma not well developed. Conidiophores, macronematous, mononematous, caespitose in groups of up to 25, emerging through stomata, pale to mid-olive brown, paler at the tip, up to 10 septate, geniculate, mostly simple, rarely branched with a conspicuous widely spaced scar of attachment at the tip. Conidia acicular, hyaline, smooth, slightly curved with a truncate base, multiseptate. The fungus was identified as *Cercospora bidentis* Tharp. This is the first record of this fungus on *H. tuberosus* fron India, although this has been reported on this host from Mauritius[1] in 1960.

We are grateful to Dr. J. E. M. Mordue of C.M.I., England, for identifying the fungus and to Dr. H. C. Govindu, Head, Department of Plant Pathology, for providing facilities.

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1. Anonymous, *Rep. Dep. Agric.*, *Mauritius*, 1960, p. 46 (1961).

Effect of Temperature on the Ecological Relationship Between *Colletotrichum falcatum* Went and a *Curvularia* Species

Extensive laminar spots were observed on green as well as dry leaves of a seven months old crop of Co. 213 on the Institute farm. This has been conclusively proved to be due to the red rot organism *Physalospora tucumanensis* Speg. (Singh and Alexander[1]). During the course of this work, several bits of the infected leaf tissues were plated on Oatmeal agar medium for isolation of the causal organism. It was observed that in spite of surface sterilisation of the leaf tissues (with 0·1% mercuric chloride solution) a species of *Curvularia* was associated with *Colletotrichum falcatum*. Both the fungi were isolated separately and brought into pure culture. It was interesting to note that at 30°C and above, *Curvularia* sp., dominated while at lower temperatures (23° C to 30° C), *Colletotrichum falcatum* was dominant. Hence, after plating the bits were incubated at 23° C and the yield was predominantly *C. falcatum*.

It can therefore be concluded that for successful isolation of red rot organism, the tissues have to be incubated at temperatures below 30° C, as otherwise the organism can be completely missed in isolations.

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Lucknow 2, *July* 12, 1976.

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1. Singh, Kishan and Alexander, K. C., *Iudian Phytopath.*, 1970, **23**, 144.

Two New Fruit Rot Diseases Caused by *Cylindrocarpon*

A survey of fruit and vegetable markets at Allahabad during 1974–75 revealed the rot of fruits of tomato (*Lycopersicon esculentum* Mill.) and guava (*Psidium guajava* L.) caused by *Cylindrocarpon tonkinense* Bugn. The pathogenicity of this fungus was confirmed on both the fruits. About 4–7% fruits got infected with this infection.

The infection on the tomato started either through the stalk end or through the skin cracks on the fruits. White cottony growth of the fungus appeared on the infected regions. The disease in the guava appeared through injury on the fruits. Subsequently, mycelium and conidia of the pathogen were evident on the lesions. As compared to guava isolates, the tomato isolates produced more chlamydospores slightly smaller in size. Earlier Nayak[1] recorded *C. tonkinense* on potato tubers but it has not been reported on tomato and guava fruits.

Grateful thanks are due to the Director, C.M.I., Kew. for confirming the identity of both the isolates (No. IMI 199030 and 199031) and to Professor K. S. Bilgrami and Dr. M. P. Tandon for guidance.

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JAMALUDDIN.

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1. Nayak, M. L., *Sci. and Cult.*, 1964, **30**, 143.

Parasitization of the Safflower Aphid by *Aphidencyrtus aphidivorus* (Meyr) (Hymenoptera : Encyrtidae)

The aphid, *Dactynotus compositae* Theobald (Syn. *Macrosiphum compositae* Theobald) is a very serious pest of safflower during *rabi* season in northern parts of Karnataka. No parasite seems to have been reported on this species of aphid.

During the months of February–March, 1975 and 1976, the authors observed the mummified aphids of *D. compositae* around Dharwar (Karnataka). They were parasitized by an endoparasite, *Aphidencyrtus aphidivorus* (Meyr). The percentage of parasitization was 5–22 during February–March, 1976. In the laboratory, the parasite was found to accept only the second instar aphids. The adult parasite was observed to rest on the aphid, make puncture with the help of ovipositor and feed on the oozing body fluid. Further, the act of oviposition was found interesting. The parasite approached the aphid, disturbed it with its antennae and followed the aphid. The parasite turned back and parasitized the aphid, while it was moving. Mummification of the parasitized aphids was observed after 4 to 5 days. The total life-cycle occupied 10 to 11 days. Longevity of the adults varied from 6·0 to 8·5 days when fed with 25% honey solution. Further studies are in progress.

The authors are thankful to Dr. S. V. Patil, Director of Instruction (Agri.), College of Agriculture, Dharwar, for his encouragement and the authorities of the British Museum, London, for identifying the parasite,

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Leaf Spot of Maize Caused by *Pithomyces maydicus* (Sacc.) Ellis in West Bengal

The occurrence of *Pithomyces maydicus* (Sacc.) Ellis on living leaves of maize at Kalimpong, West Bengal. is recorded here. Exotic varietal lines of maize grown at the State Farm, Kalimpong, were seen to be affected by a leaf spot disease during July–August, 1975. Symptoms were found generally on the younger leaves Infections, starting from the basal part, progressed towards the tip of the leaves. Brown to dark brown, 1–2 mm streaks ran along the veins. With progress of the disease, several such streaks coalesced to form a large dull brown spot of size 10–25 mm or more. Symptoms were pronounced after tasseling of the plants. Histopathological studies revealed pronounced necrosis of epidermal and mesophyll cells. Conidia on short conidiophores were clustered and emerged by rupturing the epidermis in a row. Pure cultures on PDA were faint yellow green to olive green, mycelium mostly superficial, conidia solitary, simple, detached through fractures of denticle, part of which remained attached to the base of conidia, ellipsoid to broadly ovoid or oblong, round at ends, finely echinulate with 2–8 transverse and 0–3 longitudinal septa. Conidia on host and nutrient medium measured 10–20 × 8–12 μ with average size 14·0 × 9·5 μ. The species is identified as *Pithomyces maydicus* (Sacc.) Ellis[1]. Ponappa (1967)[2] reported the occurrence of the species on *Coix lachryna-jobi* L., *Vitex negundo* L. and *Allium cepa* L. in India. This is the first record of its occurrence on maize (*Zea mays* L.) in India, causing a minor leaf spot disease. Material is deposited with the Herbarium at I.A.R.I., New Delhi-12, under Acc. No. 31998.

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April 1, 1976.

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1. Ellis, M. B., *Dematiaceous Hyphomycetes*, C.M.I, Kew, England, 1971, p. 44,
2. Ponappa, K. M., "Some interesting fungi. I. Miscellaneous fungi," *Proc. Indian Acad. Sci.*, 1966, **66** B, 262.

QUALITY OF SCIENTIFIC PUBLICATIONS IN INDIA

A Symposium on the 'Quality of Scientific Publications in India' organised by the Indian National Science Academy, New Delhi, Indian Academy of Sciences, Bangalore and the National Academy of Sciences, India, Allahabad, was held at the Indian National Science Academy premises in New Delhi on 6th and 7th October, 1976. Dr. B. P. Pal, President, Indian National Science Academy, Professor. R. N. Tandon, President, National Academy of Sciences, India and Dr. S. Ramaseshan, Vice-President, Indian Academy of Sciences, gave the opening key-note addresses.

The main objective of the Symposium was to take stock of the present status of our Scientific Publications (Journals, Books, etc.) and to consider ways of improving their standard. The participants discussed at length various aspects of Scientific Publications such as quality of scientific content, writing, editing, refereeing, printing and production. The Conference felt that a National Publication Policy for Scientific Publications should be formulated. This Committee should examine various facets related to the improvement of the quality of Scientific Publications in India including the quality of scientific content, presentation, rapidity of publication, conciseness, refereeing, editing, printing and production.

REVIEWS AND NOTICES OF BOOKS

Nuclear Structure (Vol. II)—**Nuclear Deformation.** By A. Bohr and B. Mottelson. (W. A. Benjamin, Inc., Reading, Mass.). 1975. Pp. xvii + 748. Price $ 37.50.

This book is a massive treatise on collective motion, both vibrational and rotational, and on the consequent deformations produced in nuclei. It is the second of a proposed set of 3 volumes on nuclear structure and is devoted entirely to collective motion and deformations from spherical symmetry. As is well known, Professors Aage Bohr and Ben Mottelson have themselves done pioneering work on the subject (Their years' of dedication to the field has recently received public recognition through a popular award).

Their expertise in and love of the subject-matter clearly shows in the book, which easily outruns all competition in its breadth and treatment of collective motion in nuclei. The early books by Blatt and Weisskopf (John Wiley and Sons, Inc., New York 1952) and Bethe and Morrison (John Wiley and Sons, Inc., New York, 1956) were written before most of the developments in the field took place. More recent texts like those of Preston (Addisson Wesley Publishing Co., U.S.A., 1962) or Roy and Nigam (John Wiley and Sons, Inc., New York, 1967) devote one chapter to both rotational and vibrational modes together. By contrast the present volume gives a near-700 page discussion of this subject.

Chapter four (the first in this, the second volume) is a detailed study of nuclear rotational spectra. The authors have wisely separated the case of axially symmetric nuclei from the more complicated non-symmetric ones. An attempt is made to present isotopic spin excitations as generalisation of collective rotations to iso-space. This is followed in chapter five by a study of single particle motion in non-spherical nuclei which have been deformed by collective rotation. This chapter offers detailed illustrations of the quantum mechanics of a particle, bound or scattered by non-spherically symmetric potentials, and could therefore be useful for similar problems in atomic and molecular physics. Finally, chapter six, the last and the longest, deals with vibrational modes. Apart from the usual discussion of harmonic vibration, a large section is devoted to anharmonic effects and resulting coupling of different multipole modes. Charge exchange and polarisation are treated from the interesting viewpoint of vibrations in isotopic space. The transition from vibration to fission is discussed briefly.

A more detailed description of the contents would require an order of magnitude more space. The large chapters in the book are divided into a too many sections in a conceptually disorganised sequence. We will be content with some general remarks.

Detailed and painstaking as this book is it would be too unwieldy, along with Vol. I and the proposed Vol. III, to be used as a text-book even for a one-year post-graduate nuclear physics course. Also, because of the decade or so of time that has been spent in preparing the two volumes certain major developments in nuclear structure during the past 10–15 years are covered very scantily, if at all. Examples of this are the theory of nuclear matter, cluster expansions, three-body forces and pion condensation which comprise the few conceptually new and interesting nuclear physics research areas of the past decade. Some of this work had in fact been completed well before Volume I, and there is not much excuse for their sparse treatment when the authors have set out on a massive three-volume series on Nuclear structure. One hopes that the fact that some of these topics have been briefly touched upon will not prevent them from given a more up-to-date discussion in Vol. III. Returning to this second volume under review, despite and perhaps because of its very size and level of detail, the book gives an impression of nuclear theory as a patch-work job with a different 'theory' for each occasion. To a large extent, this is not the authors' fault. Nuclei, with their uncomfortably 'intermediate' number of *strongly* interacting constituents (too many for an exact treatment and too few for most statistical methods) do not lend themselves to clean pedagogical treatment. Nevertheless, one would have expected authors of such eminence to bring about more unity amongst superficially diverse techniques.

To put things in perspective, however, this is not an easy task, and one is strongly limited by the nature of the subject-matter. The scarcity of good nuclear physics text-books, despite the 50-year history of the subject, is itself testimony to the difficulties involved. Viewed in this background, *Nuclear Structure*, Vol. II, is a giant and laudable accomplishment.

R. RAJARAMAN.

An Introduction to Pteridophyta (Diversity and Differentiation). By A. Rashid, (Vikas Publishing House Pvt. Ltd., New Delhi), 1976. Pp. xi + 283. Price Rs. 35.00.

The book under review is intended to present an integrated account of the diversity and differentiation seen in a group of plants which occupy a vantage position in the scheme of evolution of the vascular plants. Though appearing under the title of an introduction, the book has a wealth of data amassed by the author and should prove invaluable as a source of reference to the teacher and the student at the post-graduate level of instruction. It may need, however, a teacher of high competence to put across the ideas contained here to the students at the under-graduate (B.Sc.) level.

In the earlier chapters, the author has dwelt at length on the diversity of form exhibited by the representatives of the various classes of Pteridophyta, extinct as well as living, and has given a detailed exposition of the structural features of the vegetative and reproductive phases. There are numerous illustrations which are all redrawn from standard books on the subject of Eames, Foster and Gifford, Smith, Bierhorst (this name appears on every page of illustrations in this section) and others. One wishes the author had highlighted the Indian work which is quite extensive and authoritative. The remaining chapters deal with a general discussion of the gametophytes, sporophytes, sexuality, alternation of generations, etc. Recent work on the experimental aspects for which the Pteridophytes offer excellent material has received due consideration. The literature coverage is not only up to date but also includes such 'modern' approaches like electron microscope studies, surgical and other elegant experiments elucidating the morphogenic processes in ferns.

Reading through the book dealing with a group of plants having an evolutionary history dating back to nearly 500 million years, one feels that the exciting story of the plants of great beauty and which hold fascination to both amateur and professional botanists and which once dominated the world's flora with the giant lycopods and horse-tails could have been told better. As it is, a beautiful story has strayed from its charming course and run into a maze.

A little care in editing and proof reading could have avoided the many shortcomings noticed in the text. 'Selaginaceae' (p. 6), 'the stem has one-celled epidermis' (p. 22),[6] the marginal cells often form a thin extension or loose meristematic activity' (p. 165) are examples. The sub-division should end in -phytina and not in -opsida (p. 7). The consistent use of the small initial letter for the family names throughout the text is an irritant. The text is also marred by numerous errors in spelling, use of singular and plural ('stems consists'), punctuation and the reluctance to use the definite article in the appropriate places.

The book is well produced with an attractive cover and, considering the printing costs in the country, the price appears to be reasonable.

M. A. RAU.

Annual Review of Pharmacology and Toxicology. (Vol. 16). Editor: H. W. Elliott. Assoc. Editors: R. George and R. Okun. (Annual Reviews, Inc. California), 1976. Pp. 566. Price : U.S.A. $ 17·00 $ 17·50 elsewhere.

The special recognition of toxicology as an important area of pharmacology is reflected in the change in the title of the *Annual Review* to *Annual Review of Pharmcology and Toxicology.* The significant place toxicology now occupies in both therapeutic and nonmedical environmental agents is evident from the number of reviews presented in this volume. 'Toxicology of inhalation anaesthetics and metabolites ' Neurotoxic indoleamines and monoamine neurons' Current concepts about the treatment of selected poisonings', and 'Pharmacology and toxicology o lithium' deal with toxicity related to the therapeutics 'Arsenic toxicology and industrial exposure' is an account of accidental or industrial poisoning ; various methods for evaluation of pulmonary toxicity and the toxicological effects of gaseous and particulate contaminants on pulmonary microbial defense systems constitute two separate reviews. 'Behavioral pharmacology and toxicology' and 'cutaneous pharmacology and toxicology' document recent progress in these fields.

'Central nor-adrenergic control of blood pressure and 'Renin and the therapy of hypertension summarise recent concepts in the regulation of arterial pressure as also some facets of the etiology and therapy of hypertension.

The role of neural factors in the regulation of insulin and glucagon secretion, site and mechanism' of action of diuretics, the effect of melationin on Pineal gland, the therapeutic implications of Bioavilability, drug therapy of 'myopathes associated with alterations in the immune mechanisms', developmental pharmacology as related to Hepatic cytochrome P 450 monooxygenase, are some of the highly informative reviews presented in this volume.

The discussions on 'selective suppression of humoral immunity,' 'thrombogenic effect of drugs,' 'selective transport of cytoplasmic constituents in neuronal processes' illustrate the interdisciplinary nature of pharmacological research.

M. SIRSI.

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EFFECT OF WATER-METHANOL MIXED SOLVENTS ON THE ULTRASONIC RELAXATION OF CADMIUM ACETATE

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ABSTRACT

Measurements of ultrasonic absorption have been made by pulse technique in 1 M solutions of cadmium acetate with water-methanol mixed solvents. Results are analysed by assuming a single relaxation mechanism. The characteristic frequency of relaxation is found to decrease with increasing composition of methanol in the solvent. It is proposed that the mechanism of relaxation may be the perturbation of chemical equilibrium between complex Cd Ac^+ ions and Cd^{++}, Ac^- species by soundwaves.

EQUILIBRIUM chemical dissociation of an electrolyte often results in a relaxation type of ultrasonic absorption. The macroscopic dielectric constant of the solvent determines the degree of dissociation as well as the rate at which such reactions proceed in solution. In turn, varying composition of a mixed solvent must change the frequency of relaxation of a dissolved electrolyte. Systematic investigation in this direction are available in literature only for a few electrolytes. The effect of varying solvent composition on the characteristic frequency of a relaxing solute has been reviewed by Stuehr and Yeager[1] for some divalent sulphates. Krishnamurthy[2] and Murthy and Rao[3] extended these studies to some di-univalent salts. In the present note the ultrasonic absorption studies made in weak solutions of cadmium acetate with water-methanol mixed solvents are reported.

Measurements of absorption and velocity have been made by pulse technique in the range 1 to 15 MHz at room temperature. Results are analysed by assuming a single relaxation mechanism

$$\frac{\alpha}{f^2} = B + \frac{A}{1 + (f/f_r)^2}$$

where A and B are constants of relaxation, and f_r is the characteristic frequency of relaxation. The excess absorption per wavelength ($\alpha'\lambda$) is calculated from the relation

$$(\alpha'\lambda) = \left(\frac{\alpha}{f^2} - B\right) fv$$

where v is the velocity of sound.

Results of absorption are reproducible within ± 10%.

Values of ultrasonic absorption and velocity are presented in Table I together with computed parameters $\alpha'\lambda$, f_r and μ_m at different compositions of methanol in the solvent. The dependence of α/f^2 with composition of methanol is shown in Fig. 1. It can be seen from this figure that as the composition of methanol is gradually increased, α/f^2 goes through a pronounced maximum at a certain intermediate composition. In the absence of cadmium acetate in solution, no such absorption maxima could be found at intermediate compositions of water-methanol mixtures around room temperature. The existence of such maximum in α/f^2 indicates that the salt plays an important role in determining the ultrasonic behaviour of the solution.

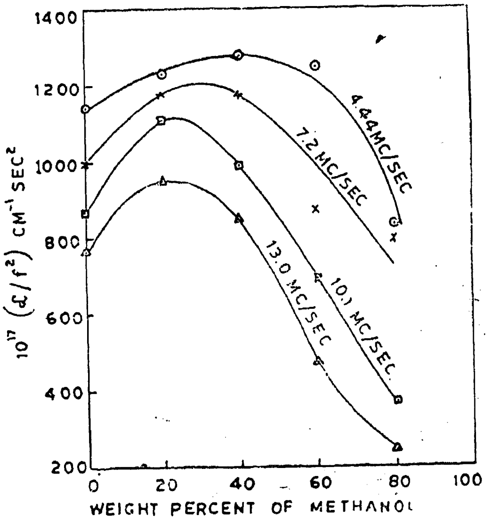

FIG. 1. Plot of α/f^2 *vs.* concentration of methanol in one molar cadmium acetate solutions.

The variation of excess absorption per wavelength ($\alpha'\lambda$) with frequency is shown in Fig. 2 for different solvent compositions. The characteristic bell-shaped absorption curves typical of relaxation appear only at 60% and 80% methanol compositions in the solvent. It appears that at lower compositions of methanol the relaxation frequency lies outside the frequency range of absorption measurements.

TABLE I

Ultrasonic absorption and relaxation parameters in one molar cadmium acetate, $3H_2O$ in water-methanol solvent mixture at 28° C

Weight% methanol	f Mc/sec.	$10^{17} \times \alpha/f^2$ cm^{-1} sec^2	$10^2 \times$ $(\alpha'\lambda)$ db	v m/sec	f_r Mc/sec
0	4·44	1135	6·3	1526	17·1
	7·20	990	9·3		
	10·10	865	11·3		
	13·00	760	12·8		
20	4·44	1230	7·4	1585	..
	7·20	1180	11·5		
	10·10	1100	15·3		
	13·00	945	16·6		
40	4·44	1275	7·2	1496	17·3
	7·20	1170	10·7		
	10·10	990	12·8		
	13·00	850	14·1		
60	4·44	1250	6·6	1398	9·3
	7·20	870	7·4		
	10·10	690	8·3		
	13·00	465	7·1		
80	4·44	925	4·7	1280	6·0
	7·20	785	6·4		
	10·10	370	4·1		
	13·00	250	4·4		

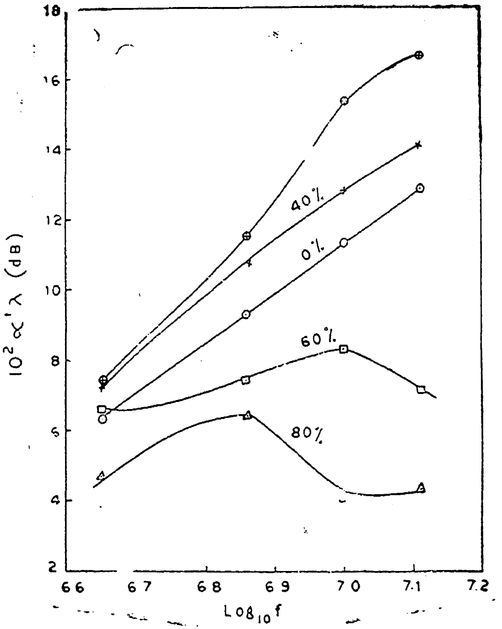

FIG. 2. Variation of $\alpha'\lambda$ with frequency in 1M solutions of cadmium acetate with water-methanol solvent mixtures.

In view of the high concentration (1 M) of cadmium acetate in solutions the mechanism of relaxation can be assumed as a perturbation of equilibrium

$$\mathrm{Cd\,Ac^+ \rightleftarrows Cd^{++} + Ac^-}$$

by sound waves. The probability for the formation of complex ion species, Cd $\mathrm{Ac^+}$, increases with decreasing dielectric constant of the solvent. Increasing methanol composition in water therefore leads to a decrease of the rate constant as well as the relaxation frequency.

ACKNOWLEDGEMENT

One of the authors (J. S. M.) wishes to record his thanks to C.S.I.R., New Delhi, for the award of a Research Fellowship.

1. Stuehr, J. and Yeager, E., *Physical Acoustics*, Vol. II, Part A, Edited by W. P. Mason, Academic Press, New York.
2. Krishnamurty, C. and Ramachandra Rao, B., *Jour. Acoust. Soc. Ind.*, 1974, **2**, 106.
3. Sree Rama Murthy, J. and Ramachandra Rao, B., *Ibid.*, 1973, **1**, 85.

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THE OBSERVER IN THE WORLD-VIEW OF PHYSICS

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ABSTRACT

The ideas introduced by the author in a couple of recent papers on entropy, irreversibility and quantum measurement are first briefly summarized. The meaning of the concept of observer introduced in these papers is developed further and implications discussed. The observer is proposed as being an entity always existing and existing in the singular as opposed to being a living system obeying laws of physics and chemistry.

CAN we, in principle, form a consistent picture of the physical world and the phenomena around us in terms of a set of laws of physics regarded as being independent of the existence or non-existence of an observer who looks at these phenomena and tries to find these laws ? An assumed positive answer to this question forms the basis and provides the motivating force for all physical sciences. In a sense, the question has always existed. For example, if one tries to derive the properties of the macroscopic world of tables and chairs from the assumed hypothesis of atoms and molecules, one such property being irreversibility, one has to take recourse to *ad hoc* operations like averaging over macroscopic lengths and time scales. These operations are justified by statements to the effect that we are macroscopic observers, our senses are too crude to sense very small distances and time scales and so on. The problem of explaining why we, as observers, are so restricted still remains. If the entire process of cognition were subject to deterministic physical laws, there would be no room for these contingent elements or randomness. The reason one is able to live with these contradictions in classical physics is that the problems can be swept under the rug by a few simple statements which are not questioned.

After the advent and the remarkable success of quantum mechanics, however, the problem of the relevance of the observer has been posed with much more seriousness. The reason for this is that 'measurement' and 'preparation of a state' are much more central concepts in quantum mechanics than these are in classical physics. The disturbance of the system as a result of measurement on a quantum system is not a small, negligible effect that can be blamed on inefficiency or carelessness; it is fundamental. The existence of states in which not all observables have precise, well-defined values at the same time even in principle, the indeterminate collapse of a system to an eigenstate of the observable being measured, etc., are features peculiar to quantum mechanics. These features lead to several well-known paradoxes, for example, the Schrodinger's cat paradox, the Einstein-Podolsky-Rosen paradox and so on.

Motivated by these problems as well as by the *ad hoc* unaesthetic and artificial nature of the assumption of an external reality existing independently of the existence of an observer, we have, in a couple of recent papers[1,2], proposed a view on the problems of entropy, irreversibility and quantum measurement in which the observer is an indispensable part of a complete description of physical reality. 'Levels of perception' are introduced as a new element in the description. A level of perception is characterized by a set of concepts and assumptions which the observer regards as *a priori*. This set determines the set of all possible states the observer can observe at the given level of perception. Any state that contradicts these concepts and assumptions cannot be observed by this observer and such states must, therefore, collapse to one of the states in the allowed set. This collapse is an irreversible process and it was proposed that such a collapse during any observation by an observer whose perception is limited is the fundamental cause for the irreversibility observed in the universe by such an observer.

It was also proposed that every set of concepts and assumptions considered *a priori* must be assigned a negentropy corresponding to the number of states considered inaccessible by virtue of these constraints. These states would become accessible for a more unrestricted observer, whose conceptual framework or 'level of perception' allows for a larger number of states. It follows that any universe seen by a restricted observer must evolve, since it has a potential negentropy. In the limiting case, an observer who is completely unrestricted and allows for all possible states sees a universe which does not evolve, for there is no potential negentropy.

The above hypotheses would clearly be of little significance if the existence of the postulated observers with various levels of perception were impossible. These would also be of little significance if these levels of perception were unattainable by 'human' observers.

It would be desirable therefore to discuss a little further what we mean by 'observer' and the relation of this observer to phenomena. Clearly, by 'observer' we mean something more than a physico-chemical system obeying fixed laws of physics and chemistry, since this is itself a classical or a macroscopic view, hence only relevant for a particular level of perception. For the same reason, it cannot be looked upon as a spatio-temporally bound system. *The observer is an entity which always exists and exists only in the singular.* There is only one observer! It is the same observer that attains these various levels of perception, including the ones in which it sees itself as many observers. The strongest argument in favour of the view is that *an observer is never experienced in the plural.* The inability to visualize such an observer is no more serious than our inability to visualize quantum mechanical objects, for example electrons. This dissociation of the concept of an observer from 'human-being' or from the 'phenomenon of life' is, we believe, important for the resolution of the age-old question: Would there be a Universe if there were no one to observe it? The answer is No, because the existence of the observer is a fundamental truth—the only truth accessible to direct experience. The concept of an external world and laws governing its phenomena are constructs invented by the observer to put order and meaning into his own experience. These constructs are, therefore, subject to change in the observer's perception, for no set of concepts meaningfully describes the entire experience.

An immediate question that arises is, whether these various levels of perception are attainable by a 'human' observer. The following way of looking at the problem may perhaps be helpful. Let us note that if we accept the more general concept of the 'observer' mentioned above, then for this observer, there is no distinction between mental phenomena like thoughts, feelings, emotions, etc., and physical phenomena like burning of wood. Both kinds of phenomena are parts of the observer's experience. The identification is useful, however, in one respect. In the realm of mental phenomena we explicitly recognize the impossibility of observation of a phenomenon without disturbing or changing the phenomenon. It is very difficult to observe thoughts without changing them. Further, mental phenomena are known to be phenomena which themselves change as a result of their understanding. For example, if repeated performance of a habitual act by a person is looked upon as a phenomenon, we do allow for the possibility that the person may get rid of the habit by conscious effort in the form of introspection. Now if there is no qualitative difference between mental and physical phenomena, both taking place in the observer's mind, one must allow for the possibility that the latter may change as a result of conscious effort on the part of the observer. Physical laws could thus be looked upon as habits, only much more deep-rooted, consequently requiring much greater effort to change.

1. Bhandari, R., *Pramāṇa*, 1974, 3, 1.
2. —, *Ibid.*, 1976, 6, 135.

SPECTROPHOTOMETRIC STUDY OF THE REACTION OF OSMIUM WITH ISONIAZID

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ABSTRACT

Isoniazid is proposed as a new reagent for the spectrophotometric determination of microgram amounts of osimum. It forms an yellow complex with Os(VIII) and Os(VI) in the pH range 3·8–4·5 at room temperature. The yellow complex exhibits maximum absorbance at 420 nm. Beer's law is valid over the concentration range 0·1–6·5 ppm for Os(VIII) and 0·4–7·0 ppm for Os(VI). The sensitivities are 0·013 μg/cm^2 for Os(VIII) and 0·026 μg/cm^2 for Os(VI). The effects of time, temperature, order of addition of reagents, reagent concentration and diverse ions are reported.

INTRODUCTION

ISONIAZID, isonicotinic acid hydrazide (INH) is extensively used in the treatment of tuberculosis. It has been proposed for the spectrophotometric determination of vanadium[1] and gold[2]. In the present paper the authors have investigated the colour reaction between INH and the platinum metals and developed INH as a sensitive reagent for the spectrophotometric determination of osmium. The proposed method offers the advantages of simplicity and good sensitivity without the need for extraction.

TABLE I

Determination of osmium in synthetic mixtures corresponding to syserkite

Osmium (VIII) taken (ppm)	Ir (ppm)	Ru (ppm)	Pt (ppm)	Rh (ppm)	Au (ppm)	Osmium (VIII) found (ppm)
0·8	0·5	0·25	0·20	0·05	0·01	0·85
1·0	0·9	0·40	0·40	0·01	0·05	1·03
1·5	1·2	0·60	0·50	0·03	0·10	1·51
2·4	2·0	0·80	0·60	0·02	0·10	2·42
3·0	2·5	0·50	0·10	0·01	..	3·04
5·0	4·0	0·80	0·20	0·01	..	5·00

EXPERIMENTAL

Standard solution of osmium (VIII) was prepared from osmium tetroxide (M/s. Johnson Matthey Chemicals Ltd., London) in 0·02 M sodium hydroxide. Standard solution of osmium (VI) was prepared from potassium osmate (M/s. JMC) in doubly distilled water. Buffer solutions of the pH range 0·65–5·2 were prepared from 1 M sodium acetate and 1 M hydrochloric acid solutions. A 1% aqueous solution of INH was used. Standard solutions of diverse ions were prepared from analytical grade chemicals. Beckman Model DB spectrophotometer was used for absorbance measurements.

Procedure :—The sample solution containing 20–150 μg of osmium (VIII) or 25–175 μg of osmium (VI) was transferred to a 25 ml volumetric flask. 5 ml of Walpole buffer of pH 3·6 and 5 ml of 1% INH were added and diluted to the mark with doubly distilled water. After 10 minutes, the absorbance was measured at 420 nm against a reagent blank prepared under similar condition. The osmium concentration of the sample solution was then deduced from the standard calibration curve.

RESULTS AND DISCUSSION

Of the six platinum metals only octavalent and sexavalent osmium forms an yellow coloured complex with INH at room temperature (27° C) in the presence of Walpole buffer. Osmium (VI) does not form any coloured complex with INH. Maximum colouration takes place in the pH range 3·8–4·5. All studies are therefore confined to pH 4. The yellow colour is fully developed in 10 minutes after mixing the reagent and is stable for 10 minutes. The order of mixing osmium (M), buffer (B) and INH (L) has some effect on the development and stability of the colour. If the mixing of the reagents follows the order : MBL, BML or LBM, the maximum colour development takes place in 10 minutes. If the mixing of the reagents follows the order : LMB or MLB the full development of colour takes place in 15 minutes. It is therefore recommended to mix osmium and buffer solutions prior to the addition of INH solution.

The minimum amount of INH required for the maximum development of colour is 5 ml of 1% solution. Higher concentration of the reagent does not alter the colour intensity. The absorbance readings are constant in the temperature range 10–45° C.

Beer's law is obeyed over the concentration range 0·1–6·5 ppm of osmium (VIII) and 0·4–7·0 ppm of osmium (VI). The optimum concentration ranges as evaluated by Ringbom plot are 0·8–6·0 ppm for osmium (VIII) and 1·0–7·0 ppm for osmium (VI). According to Sandell's expression, the sensitivity of the reaction is 0·013 μg/cm² for osmium (VIII) and 0·026 μg/cm² for osmium (VI). The absorbance of six solutions containing 5 ppm of osmium has a maximum relative deviation of 1·5% and an average deviation of 0·5%.

The sensitivity of the proposed method is more than that of *o*-(β-benzoylthioureido) benzoic acid[3], 2-mercaptobenzimidazole[4], 2-mercaptobenzothiazole[5], 1-carbamidino-3-methyl-5-pyrazolene[6], bismuthiol II[7] and β-benzoyl-α (*o*-tolyl) thiourea[8] which have been proposed as sensitive spectrophotometric reagents for osmium (VIII).

The yellow colour of the solution is assumed to be the colour of a complex because the solutions of the reagents and the oxidation products of INH are colourless. Job's method of continuous variation[9,10], mole ratio method[11] and limiting logarithmic method[12] fail to give the composition of the complex because the yellow colour is not developed even in the presence of 200-fold molar excess of INH. A 925-fold molar excess of INH is necessary for the complete complex formation. The cationic nature of the complex is indicated by the retention of the yellow colour on Dowex 50W–X8 cation exchange resin.

The following amounts (μg/ml) of foreign ions are found to give less than 2% error in the determination of 3 μg/ml osmium: Pd^{2+} (20), Pt^{4+} (1·0), Ru^{3+} (1·0), Rh^{3+} (8·0), Ir^{3+} (8·0), Cu^{2+} (16·0), Ni^{2+} (400), Co^{2+} (44), Au^{3+} (0·2), Fe^{3+} (100), Ag^{+} (0·4), Mn^{2+} (80), F^{-} (1400), Cl^{-} (1600), Br^{-} (600), I^{-} (600), SO_4^{2-} (1000), NO_3^{-} (1600), PO_4^{3-} (400), $S_2O_3^{2-}$ (20), EDTA (80), acetate (800), oxalate (12) and citrate (8). Pt^{4+} and Ru^{3+} interfere because of their colour.

Determination of osmium in ores.—Synthetic mixtures corresponding to syserkite were prepared and the osmium content was determined following the standard procedure. The results are given in Table I.

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1. Zdzislaw Krych. and Tadeusz Lipiec, *Chem. Anal.* (*Warsaw*), 1967, 12, 535.
2. Adil, A. S., Ayaz, A. A., Ahmed, I. and Hasmi, M. H., *Mikrochim. Acta*, 1970, 3, 606.
3. Majumdar, A. K. and Bhowal, S. K., *Anal. Chim. Acta*, 1966, 35, 479.
4. Bera, B. C. and Chakrabartty, M. M., *Anal. Chem.*, 1966, 38, 1419.
5. — and —, *Microchem. J.*, 1966, 11, 420.
6. Poddar, S. N. and Adhya, J. N., *Sci. & Cult.* (*Calcutta*), 1964, 30, 50.
7. Majumdar, A. K. and Bhowal, S. K., *Anal. Chim. Acta*, 1972, 44, 157.
8. Bhowal, S. K., *Curr. Sci.*, 1975, 44, 157.
9. Job, P., *Ann. Chim.*, 1928, 9, 113.
10. Vosburg, W. C. and Copper, G. R., *J. Amer. Chem. Soc.*, 1941, 63, 437.
11. Yoe, J. H. and Jones, A. L., *Ind. Eng. Chem., Anal. Ed.*, 1944, 16, 111.
12. Bent. H. E. and French, *J. Amer. Chem. Soc.*, 1941, 63, 568.

PROTON-LIGAND FORMATION CONSTANTS AND FORMATION CONSTANTS OF Tl (I)-3-NITRO SALICYLATES

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ABSTRACT

Proton-ligand formation constants and formation constants of Tl (I)-complexes with 3-nitro salicylie acid have been determined at 30° C (50% aqueous ethanol, 0·1 M $NaClO_4$) by various computational methods. The average values of proton-ligand formation constants are found to be $\log pK_1^H = 10·34$ and $\log pK_2^H = 2·30$, the corresponding values for formation constants are 7·87 and 5·05 respectively.

ALTHOUGH the co-ordination chemistry of salicylic acid is well documented, the study of its derivative, 3-nitro salicylic acid (3-NSA), remained neglected until quite recently. Khadikar *et al.*[5-7] have reported the complex forming tendency of 3-NSA with number of di- and tri-valent metal ions. The literature does not reveal the complex formation of Tl(I) with 3-NSA. The present study deals with the same. The proton-ligand formation constants of 3-NSA and the formation constants of its complexes with Tl(I) have been determined in the present investigation employing half-integral, point-wise calculation, and linear plot method. Refined values of formation constants are obtained by the method of least-squares.

EXPERIMENTAL

All the chemicals used were of Analar B.D.H. grade.

Polymetron model CL–41 was used for pH titrations conducted at 30°. Measurements were made with an accuracy of ± 0·05 pH units and the reproducibility of the readings was of the same order. The pH-meter was calibrated using Cambridge buffer tablets of pH 4·1 and 9·1.

pH-metric titrations of solutions of (*i*) free $HClO_4$, (*ii*) free $HClO_4$ + 3-NSA and (*iii*) free $HClO_4$ + 3-NSA + Tl(I), were performed in 50% (v/v) aqueous ethanol medium against standard NaOH solution while maintaining the ionic strength at 0·1 M $NaClO_4$.

TABLE I

pH	$\bar{n}_A$	$\log \frac{\bar{n}_A}{1-\bar{n}_A}$	$\log \frac{\bar{n}_A - 1}{2-\bar{n}_A}$	Point-wise	
				$\log pK_1^H$	$\log pK_2^H$
2·2	1·614		+0·201		2·401
2·3	1·518		+0·031		2·331
2·4	1·449		−0·089		2·311
2·6	1·259		−0·456		2·144
2·8	1·156		−0·733		2·067
3·0	1·017				
4·0	0·992				
5·0	0·985				
6·0	0·980				
8·0	0·975				
9·0	0·945				
9·8	0·831	+0·691		10·491	
10·0	0·742	+0·459		10·459	
10·1	0·706	+0·381		10·481	
10·2	0·623	+0·219		10·419	
10·3	0·472	−0·048		10·252	
10·4	0·452	−0·082		10·318	
10·5	0·327	−0·311		10·189	
10·6	0·277	−0·416		10·184	

The experimental set-up and the method of calculation is the same as described earlier[1-3].

RESULTS AND DISCUSSION

The Irving-Rossotti expression[4] was used for the calculation of $\bar{n}_A$ (Degree of formation of proton-ligand complex), $\bar{n}$ [Degree of formation of Tl(I)-complex], and $p(L)$ (Free ligand exponent). The cocentrations were corrected for the changes in volume as a result of addition of alkali during titration. A series of values of $\bar{n}_p$, $\bar{n}$ and $p(L)$ corresponding to different B-values (pH-meter readings) were calculated (Tables I-II) and the formattion curves for ligand and complex were obtained by plotting $\bar{n}_A$ *vs.* pH and $\bar{n}$ *vs.* $p(L)$ respectively (Figs. 1–2). The values of log pK_1^H, log pK_2^H (Practical proton-ligand formation constants) and also log K_1 and log K_2 were recorded directly from the respective formation curves from integral values of $\bar{n}_A$ and $\bar{n}$ and are recorded in Table III. Values of these constants were also obtained by point-wise calculation as well as linear plot method[1] (Figs. 3–4). Refined values of log K_1 and log K_2 were obtained by the method of least-squares.

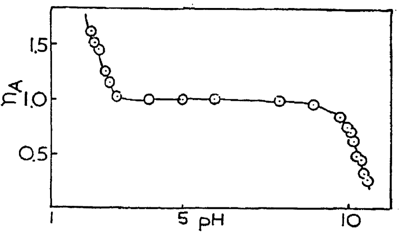

FIG. 1

Persual of Table II (Fig. 2) shows that $\bar{n}$ value never exceeds 1·7 indicating thereby the formation

TABLE II

$\bar{n}_A$	$\bar{n}$	$\log \frac{\bar{n}}{1-\bar{n}}$	$\log \frac{\bar{n}-1}{2-\bar{n}}$	p(L)	Point-wise log K_1	Point-wise log K_2
0·985	0·158	−0·458		8·798	8·340	
0·983	0·310	−0·345		8·567	8·222	
0·984	0·453	−0·08		8·034	8·954	
0·985	0·670	+0·308		7·382	7·690	
0·983	0·723	+0·417		7·132	7·549	
0·981	0·778	+0·547		6·902	7·449	
0·982	1·086		−1·026	5·590		4·564
0·978	1·148		−0·760	5·480		4·720
0·975	1·365		−0·240	5·230		4·990
0·974	1·525		+0·043	5·112		4·155
0·972	1·637		+0·244	4·997		3·241
0·970	1·750		+0·477	4·877		4·354

TABLE III

Method	log pK_1^H	log pK_2^H	log K_1	log pK_2
Half-integral	10·35	2·30	7·91	5·12
Linear-plot	10·32	2·35	7·90	5·12
Point-wise calc.	10·35	2·25	7·86	5·00
Least-square	..	..	7·80	5·01
Average	10·34	2·30	7·87	5·05

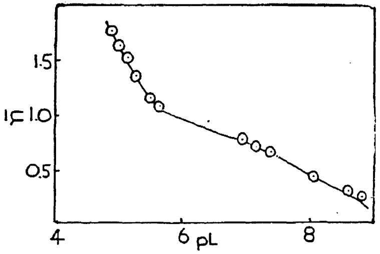

FIG. 2

of only 1 : 1 and 1 : 2 complexes of Tl(I) with 3-NSA. In view of the very low concentration of Tl(I) (1×10^{-4}) used in the titration, formation of polynuclear complexes may be ignored. Titrations performed with different ratios of metal to ligand showed that the formation constants are independent of metal ion concentration.

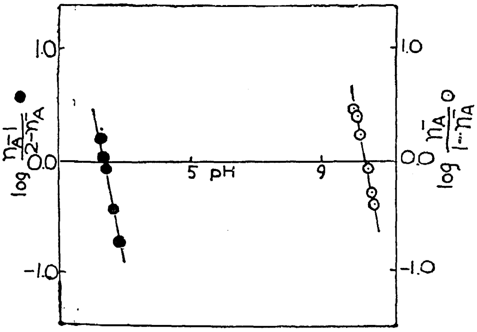

FIG. 3

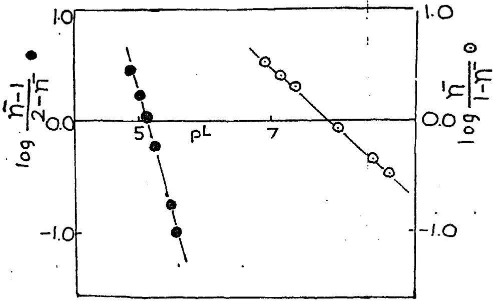

FIG. 4

The ratio of log K_1/log K_2 is positive and that the separation factor between the first and the second formation constant is well within the range. A bigger difference between log K_1 and log K_2 is due to the possible steric hindrance on the linking of the second ligand molecule expected for Tl(I) ion.

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1. Khadikar, P. V. and Ameria, R. L., *J. Indian Chem. Soc.*, 1973, 50, 389.
2. —, Poonia, N. S. and Kakkar, S. N., *Indian J. Chem.*, 1973, 11, 709.
3. — and Ameria, R. L., *J. Indian Chem. Soc.*, 1972, 49, 1049.
4. Irving, H. M. and Rossotti, H. S., *J. Chem. Soc.*, 1954, p. 2904.
5. Khadikar, P. V. and Ameria, R. L., *J. Indian Chem. Soc.*, 1973, 50, 513.
6. — and —, *Ibid.*, 1972, 49, 33.
7. — and Mehrotra, S. N., (Unpublished results).

SPECTROPHOTOMETRIC INVESTIGATION OF Ti(IV) COMPLEXES OF N-SALICYL PHENYL HYDROXYLAMINE

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ABSTRACT

Yellow coloured complexes of Ti(IV) and N-salicyl phenyl hydroxylamine are extractable into isoamyl alcohol. The spectrophotometric studies following Job's and molar ratio methods have shown that 1 : 2 and 1 : 3 (metal: ligand) complexes predominate in the acidity ranges of 2–5 N and 7·5–9 N HCl respectively. The values of the stability constants, stepwise and overall, have been calculated following extended Yatsimirskii's, Leden's and Harvey-Manning's methods. An electronic computer, IBM 1130, using FORTRAN Programming was utilized for evaluating stability constants of 1 : 3 system. The analytical suitability of the reagent for microdetermination of titanium has also been investigated.

INTRODUCTION

THE application of the aryl hydroxamic acids and their N-phenyl analogues are well documented[1-5]. But in many cases[6,7], investigations were carried out to study the complexes with the variation of acidity. The present paper deals with the spectrophotometric investigation of Ti(IV) complexes of N-salicyl phenyl hydroxylamine (NSPHA) at varied acidities. The study has established the existence of 1 : 2 and 1 : 3 (metal : ligand) complexes depending on the pH. The stepwise and overall formation constants are evaluated by extended Yatsimirskii's[8], Leden's[9] and Harvey-Manning's[10] method. The analytical suitability of the reagent for microdetermination of titanium is explored in the light of above observations.

MATERIALS

A stock solution of titanium (2·18 mg/ml) was prepared from potassium titanyl oxalate (A.R.) and was standardised[11]. The reagent, NSPHA (m.p. 106–107° C), was prepared following the method of Shome[12]. Chemicals used in all spectrophotometric measurements were of A.R. or spectrograde quality. A Hilger Uvispek spectrophotometer was used for spectrophotometric measurements. For evaluation of the formation constants, a IBM 1130 scientific and electronic computer based on FORTRAN Programming was employed.

EXPERIMENTAL

An aliquot of titanium solution ($8 \cdot 352 \times 10^{-5}$ M) was taken in a 100 ml separatory funnel and its pH adjusted to the desired value. The reagent solution (0·087 M) in isoamyl alcohol was then added and the mixture shaken thoroughly for 10 minutes. The yellow coloured nonaqueous layer was collected in a small beaker. The extraction was repeated with a 5 ml portions of isoamyl alcohol. The combined extract, after drying over sodium sulphate, was diluted to 25 ml with isoamyl alcohol and its absorption was measured at 390 and 400 nm.

RESULTS AND DISCUSSION

The absorbance measurements of the complex solution at 8 N HCl show maximum absorbance at 370 nm. Due to low reagent absorption around 390–400 nm, all the absorption measurements were taken against the reagent blank at

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* A portion of this work was included in the Ph.D. Thesis submitted to Jadavpur University in 1974.

TABLE I

Stepwise stability constants of titanium complexes at 27 ± 2° C

Method	For 1 : 2 (Metal : Ligand)			For 1 : 3			
	log K_1	log K_2	log β_2	log K_1	log K_2	log K_3	log β_3
Yatsimirskii's	2·29	2·43	4·72	2·15	1·49	3·05	6·69
Leden's	1·28	2·89	4·17	2·13	2·04	3·17	7·34
Harvey-Manning's	..	..	4·34	..	..	..	7·57

400 nm with 8 N HCl. Extraction with 6 ml of reagent was adequate for quantitative extraction with 8 N HCl. Measurements of absorbance of different sets of complementary solutions (both Job's and molar ratio methods) at 390 and 400 nm show that 1 : 2 and 1 : 3 (metal : ligand) complexes predominate in the acidity ranges of 2–5 N and 7·5–9 N HCl respectively. The systems obeyed Beer's law over the concentration ranges 1–15 ppm with 8 N HCl. The optimum concentration ranges, evaluated from Ringbom's curves, were found to be 5–14 ppm at 8 N HCl. The relative analysis error per one per cent absolute photometric error, following Ayre's equation, was found to be of the order of 2·72. The molar absorptivity of the complexes, calculated from Beer's law data at 8 N (400 nm), is (1·9 ± 0·02) × 10^3 l.mole^{-1} cm^{-1}. The sensitivity, according to Sandell, is 0·021 μg with 8 N HCl.

Moderate amounts of ions, commonly associated with titanium, did not interfere with the estimation. Fe(III), V(V), Cr(VI), Mo(VI) and U(VI) however interfere in the estimation. The reagent can therefore be used for microdetermination of titanium.

The stepwise formation constants of 1 : 2 and 1 : 3 systems were determined in the manner stated earlier[13]. Following Yatsimirskii's method, stepwise formation constants (K_1, K_2 and K_3) of 1 : 3 system were evaluated after solving a fourth degree polynomial equation in K_2 by a scientific and electronic computer (IBM 1130) using FORTRAN Programming and the results are given in Table I.

In the light of the data given in Table I, the formula of 1 : 2 complex may be represented as [$TiOR_2$] where HR stands for NSPHA.

The higher value of K_3 for the 1 : 3 complex may be due to the formation of internally hydrogen bonded 1 : 3 chelate system.

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1. Alimarin, I. P., Sudakov, F. P. and Golovkin, B. G., *Russian Chemical Reviews*, 1962, 31, 466.
2. Majumdar, A. K., *N-Benzoyl Phenyl Hydroxylamine and its Analogues*, Pergamon Press, Oxford, 1972.
3. Bhura, D. C., *N-Phenyl Benzohydroxamic Acid and Analogues as Analytical Reagents*, Technical News Service, (Sarabhai M. Chemicals), 1973, Parts 1 and 2, Vol. 5, Nos. 3 and 4.
4. Dutt, R. L., *J. Indian Chem. Soc.*, 1957, 34, 311.
5. Ghosh, N. N. and Bhattacharya, A., *Ibid.*, 1967, 44, 972.
6. Brandt, W. W., *Rec. Chem. Prog.*, 1960, 21, 159.
7. Springer, V. and Benedictov, I., *Chem. Zvesti.*, 1965, 19, 481.
8. Yatsimirskii, K. B. and Fedorova, T. I., *Zhur. Neorg. Khim.*, 1956, 1, 2310.
9. Leden, I., *Z. Phys. Chem.*, 1941, 188, 160.
10. Harvey, A. E. and Manning, D. L., *J. Am. Chem. Soc.*, 1950, 72, 4488 ; 1952, 74, 4744.
11. Vogel, A. I., *Quantitative Inorg. Analysis*, Longman's, Green and Co. Ltd., London, 1960, 3rd Ed., p. 790.
12. Shome, S. C., *Analyst*, 1950, 75, 27.
13. Chakrabarti, A. K. and Bag, S. P., *Z. Anal. Chem.*, 1973, 265, 259 ; 1974, 272, 124.

VII INTERNATIONAL CONGRESS ON RHEOLOGY

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The Seventh International Congress on Rheology was held in Gothenburg, Sweden, at the Chalmers University of Technology from August 23 to 27, 1976, under the Presidentship of R. S. Marvin of U.S.A. The Congress was sponsored by 29 organisations including the Swedish Board of Technical Development. 775 participants from 25 countries registered themselves for the Congress. Prof. B. R. Seth from India presented a paper on the opening day on 'Generalized Measures in Rheology' and also presided over one of the technical sessions. Three papers presented at the Congress were based on his work. In all 468 papers were communicated in five parallel sessions. The topics dealt with included theory, fluids, polymers (melts, solids, solution, elastomers), food, suspensions, concrete and metals.

The participants were welcomed by H. Hansson, President of the City Council, S. Olving, Rector of Chalmers University of Technology and F. K. G. Odquist who paid glowing tribute to late M. Reiner, for his extensive contributions to Rheology. R. S. Marvin gave a brief history of the International Committee on Rheology and H. Markovitz gave the Boltzmann centenary address on "Boltzmann and his superposition principle" in relation to the formulation of constitutive equations for linear viscoelasticity.

A number of general addresses and discussions took place, and the participants were: J. G. Oldroyd, G. Astarita, A. S. Lodge, J. Meissner, G. V. Vingradov, F. H. Muller, D. R. Axelrad. Oldroyd, Astarita, Lodge, Meissner, Vinogradov, Muller, and Axelrad took prominant part. W. V. Chang, R. Bloch, N. W. Tschoegl and M. T. Shaw, by using Seth's generalised measure, explained experimental results on the extensional viscosity of polythene melts and other rheological materials.

The new concept of Rheo-Genetics was introduced by L. Dintenfass to show that significant differences exist in the blood rheology of cigarette smokers and non-smokers. It is found that smoking gives rise to elevated concentration of red cells, blood viscosity and plasma viscosity. There were a number of social functions including the Congress Banquet in the famous Langedrag restaurant. The Congress was highly educative and the delegates evinced a lot of interest in the various items of presentation. The next Congress in 1980 will be held in Italy.

ANNOUNCEMENTS

AWARD OF RESEARCH DEGREES

Karnatak University, Dharwar, has awarded the Ph.D. degree in Chemistry to Shri Kakhandki Lachchappa Shivalingappa and Ph.D. degree in Botany to Shri Koppar Keshav.

The M.S. University of Baroda has awarded the Ph.D. degree in Physics to Shri John K. Wassly; Ph.D. degree in Chemistry to Shri Bharatkumar Trikamal Thaker,

Marathwada University, Aurangabad, has awarded the Ph.D. degree in Botany to Shri Arunkumar M. Nahemiah.

Osmania University, Hyderabad, has awarded the Ph.D. degree in Physics to Kumari K. G. Subadra; Ph.D. degree in Chemistry to Smt. Sushama Kandlikar.

Shri Venkateswara University, Tirupati, has awarded the Ph.D. degree in Chemistry to Shri K. V. Krishna Rao.

Utkal University, Bhubaneswar, has awarded the Ph.D. degree in Chemistry to Shri Umesh Chandra Mishra and Shri V. O. Kuruvila; Ph.D. degree in Anthropology to Miss Besanti Rath.

University of Cochin has awarded the Ph.D. degree in Marine Sciences to Shri P. P. Varghese.

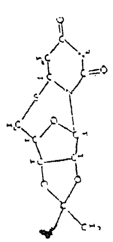
LETTERS TO THE EDITOR

CRYSTAL DATA ON THIONUCLEOSIDES

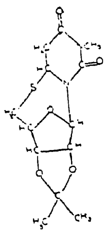
We wish to report here the crystal data of the following thionucleosides :

(1) 5′-deoxy-5′, 6 epithio-5, 6-dihydro-2′, 3′-O-isopropylidene uridine (I)

(2) 5′-deoxy-5′-6 epithio-5, 6-dihydro-N-methyl-2′, 3′-O-isopropylidene uridine (II)

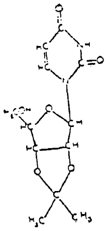


$C_{12}H_{16}N_2O_5S$
(I)



II

$C_{13}H_{18}N_2O_5S$
(II)



III

$C_{12}H_{16}O_6N_2$
(III)

Compounds I and II are purely synthetic and are of interest in connection with the work being done to provide an explanation for how substituents like CH_3 are put on to C–5 position (Private communication from Dr. D. M. Brown, University of Cambridge, U.K.). There are many minor nucleosides in *t*RNA as well as in DNA with C–5 substituents. It is important to know whether the sulphur atom is axial or equatorial on the dihydrouracil ring. We also report here the crystal data of 2′, 3′-O-isopropylidene uridine (III), an important intermediate in the preparation of many 5′-substituted uridines.

In crystallizing compounds (I) and (II), about 3 mg of the substance was dissolved in 1 ml of acetone and the solution was layered on a column of distilled water (about 4 ml) in a suitable test tube and allowed to diffuse slowly. Crystals appeared either at the interface or on the top of the acetone column in about 10 to 15 days.

Compound (I) crystallized as needles (about 2 mm long, 0·5 mm cross-section). Compound (II) crystallized as thin plates (about 1·5 × 1 × 0·1 mm). Compound (III) was crystallized by slow evaporation of its solution containing 50% acetone and 50% water. All the crystals are found to be stable in air.

The unit cell dimensions and the space groups of the crystals were determined from rotation, Weissenberg and precession photographs. They belong to orthorhombic system. The densities of (I) and (III) were determined by flotation method using chlorobenzene and bromobenzene. Crystal data are summarised in Table I.

TABLE I

	Crystal I	Crystal II	Crystal III
a	36·129 Å	39·526 Å	19·846 Å
b	14·664 Å	6·607 Å	12·774 Å
c	5·469 Å	5·661 Å	5·222 Å
Volume	2897·45 Å^3	1478·36 Å^3	1323·77 Å^3
Systematic absences	$h00: h = 2n + 1$	$h00: h = 2n + 1$	$h00: h = 2n + 1$
	$0k0: k = 2n + 1$	$0k0: k = 2n + 1$	$0k0: k = 2n + 1$
		$00l: l = 2n + 1$	$00l: l = 2n + 1$
Space group	$P2_12_12$	$P2_12_12_1$	$P2_12_12_1$
Z	8	4	4
ρ_{obs}	1·39 gm/cc	No suitable liquid for experimental determination	1·41 gm/cc.
ρ_{cal}	1·40 gm/cc.	1·44 (?) gm/cc	1·40 gm/cc

We thank Drs. D. M. Brown and Steve Salisbury, University of Cambridge, U.K., for providing the substances.

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RELATION BETWEEN FORCE FIELD PARAMETER AND MASS RATIO

THE fundamental problem of intra-molecular mechanics is the determination of the normal co-ordinate transformation matrix L which may be written as L = TA, normalised to the inverse kinetic energy matrix G in the sense $T\tilde{T} = G$. A is an orthogonal matrix. For a two-dimensional vibrational species, the A matrix can be generated by a single parameter *c*. From studies conducted on isotopic species[1] it is clear that the parameter *c* is mass-dependent, but the true functional form of this dependence cannot be determined from the theory of molecular vibrations. Müller and collaborators[2] have investigated the variation of the ratio L_{12}/L_{21} with the mass coupling parameter defined as $T = G_{12}/|G|^{\frac{1}{2}}$ which turns out to be a function of masses and geometry of the molecule and have noted certain regularities. In the present note we have, however, studied the variation of the parameter *c* with the mass-ratio m_y/m_x in a number of molecules of the XY_2 and XY_4 types.

TABLE I

Values of mass-ratio and parameter

Molecular Type	Molecule	Mass ratio m_x/m_y	Parameter *c*	Reference for exact force fields
XY_2	SeO_2	0·203	0·046	3
	ClO_2	0·451	0·082	4
	SO_2	0·499	0·084	5
	OF_2	1·188	1·156	6
	OCl_2	2·216	0·256	7
XY_4	GeF_4	0·262	−0·048	8
	$GeCl_4$	0·469	−0·013	8
	VCl_4	0·696	0·02	8
	$TiCl_4$	0·740	0·022	8
	CF_4	1·582	0·15	8

TABLE II

Force fields and coriolis constants for XY_2 and XY_4 type molecules*

XY_2				XY_4						
	Force fields (A_1)				Force fields (F_2)			Coriolis constants ζ_4		
Molecule	Present Work	Previous Result	Reference	Molecule	Present Work	Previous Result	Reference	Present-Work	Previous Result	Reference
NO_2	12·375 0·390 1·188	12·309 0·592 1·100	9	SiF_4	6·592 0·345 0·435	6·406±0·37 0·291±0·15 0·438±0·01	8	0·506	0·56	14
Cl_2S	2·586 −0·021 0·295	2·58 0 0·294	10	$SiCl_4$	2·744 0·109 0·242	2·96 ±0·09 0·14 ±0·03 0·236±0·005	8	−0·361	−0·23± 0·05	15
MgF_2	2·236 −0·02 0·1465	2·321 0·03 0·141	11	$SnCl_4$	2·530 0·121 0·122	2·688±0·08 0·22 ±0·1 0·118±0·01	8	0·284	0·27± 0·05	16
SiF_2	5·0398 −0·063 0·362	5·329 0·174 0·44	12	RuO_4	6·699 0·369 0·390	6·49 ±0·05 0·07 ±0·05 0·381±0·05	8	0·420	0·32	17
CaF_2	2·364 0·014 0·085	2·48 0·05 0·08	13	CsO_4	7·903 0·358 0·441	8·11 ±0·08 0·1 ±0·1 0·47 ±0·01	8	0·507	0·407	17

* The three values of the force constants reported correspond to F_{11}, F_{12}, and F_{22} in the case of XY_2 type molecules and F_{33}, F_{34} and in the case of XY_4 type molecules,

The parameter c has been evaluated for XY_2 type molecules like SeO_2, ClO_2, SO_2, OF_2 and OCl_2 and for XY_4 type molecules like GeF_4, $GeCl_4$, VCl_4, $TiCl_4$ and CF_4 using published force fields based on vibrational frequencies and other experimental data. Plots of c versus m_y/m_x have been drawn and are found to be linear (Fig. 1). Values of c as well as m_y/m_x are collected in Table I.

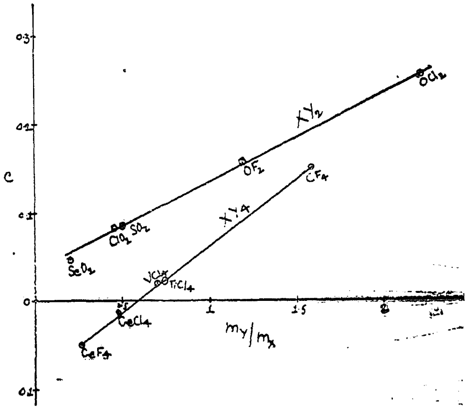

FIG. 1. Plots of parameter c *vs*. mass-ratio m_y/m_x.

With the help of the parameter-mass-ratio graphs an attempt has been made to determine the force fields and Coriolis constants of several XY_2 and XY_4 type molecules. The results of the investigation (Table II) are seen to be in good agreement with force fields and experimental Coriolis constants reported in the literature. In the light of the results reported here, it is highly plausible that smooth relationships may exist between the parameter and mass-ratio in other XY_n systems also. Such relationships will be of value in the elimination of spurious force fields and the correct assignment of frequencies. These aspects of the problem are being looked into.

One of the authors (AJV) is grateful to the CSIR, New Delhi, for the award of a Junior Research Fellowship.

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1. Ananthakrishnan, T. R., Girijavallabhan, C. P. and Aruldas, G., *J. Mol. Structure*, 1973, 16, 149.
2. Müller, A., Mohan, N. and Heidborn, V., *Z. Naturforsch.*, 1972, *27 a*, 129.
3. Takeo, H., Hirota, E. and Morino, Y., *J. Mol. Spectrosc.*, 1970, 34, 370.
4. Krishna Pillai, M. G. and Curl (Jr.), R. F., *J. Chem. Phys.*, 1962, 37, 2924.
5. Morino, Y., Kikuchi, Y., Saito, S. and Hirota, E., *J. Mol. Spectrosc.*, 1964, 13, 95.
6. — and Saito, S., *Ibid.*, 1966, 19, 435.
7. Rochkind, M. M. and Pimentel, G. C., *J. Chem. Phys.*, 1965, 42, 1361.
8. For Original reference see Mohan, N., Schmidt, K. H. and Müller, A., *J. Mol. Structure*, 1972, 13, 155.
9. Arakawa, E. T. and Nielsen, A. H., *J. Mol. Spectrosc.*, 1958, 2, 413.
10. Stammreich, H., Forneris, R. and Sone, K., *J. Chem. Phys.*, 1955, 23, 972.
11. Mann, D. E., Calder, G. V., Seshadri, K. S., White, D. and Linevsky, M. J., *Ibid.*, 1967, 46, 1138.
12. Khanna, V. M., Hauge, R., Curl (Jr.), R. F. and Margrave, J. L., *Ibid.*, 1967, 47, 5031.
13. Calder, V., Mann, D. E., Seshadri, K. S., Allavena, M. and White, D., *Ibid.*, 1969, 51, 2093.
14. McKean, D. C., *Spectrochim Acta*, 1966, 22, 269.
15. Bürger, H, and Ruoff, A., *Ibid.*, 1968, 24A, 1863.
16. Müller, A. and Königer, F., *J. Mol. Structure*, 1976, 30, 195.
17. —, Schmidt, K. H. and Mohan, N., *J. Chem. Phys.*, 1972, 57, 1752.

ON THE Z-DEPENDENCE OF TOTAL TO K-SHELL PHOTOELECTRIC CROSS SECTION RATIOS AT K-THRESHOLD

THERE have been very few studies on the total to K-shell photoelectric cross section ratios (T/K) at the K-edges. In the absence of reliable theoretical predictions, till recently, some empirical thumb rule of '5/4' law was used[1]. Later, Davisson[2] deduced the values of the total to K-shell ratios from the data of Kirchner for the first time systematically. From such deduced data Z-dependence of the ratio is found. The value is found to vary from 1·09 for aluminum to 1·235 for uranium. Based on the measurements of such ratios, empirical expressions were proposed by Allen[3] and Hubbell[4] to express the Z-dependence. However, recent theoretical[5,6] as well as experimental[7,8] data at higher energies show that deduced values at K-threshold are under estimated specially in high Z-elements. Hence, it is of interest to measure and study the Z-dependence of these ratios at K-threshold. In the recent investigations in these laboratories, the total photon attenuation coefficients above and below the K-edges are measured in the elements Cu, Zr, Ag, Sn, Ta, Pb, Th and U using proportional counters and Ge (Li) systems on a good geometry set-up[9,10]. Using these attenuation coefficients the photoelectric cross sections are deduced by subtracting the coherent and incoherent scattering contributions. The deduced photoelectric cross sections are fitted to log-log curves using IBM 1130 computer facility and the total to K-shell photoelectric cross section ratios at the respective K-edges in the elements are deduced. In the present note, a study of the Z-dependence of these ratios is made using the measured values.

The measured ratios are fitted to a polynomial expression using the IBM 1130 computer. The best fit is obtained as :

$$T/K = 1 + 7{\cdot}819 \times 10^{-3} Z - 1{\cdot}8 \times 10^{-4} Z^2 + 7{\cdot}625 \times 10^{-7} Z^3.$$

The values obtained from this expression are plotted in Fig. 1 alongwith those for the expressions of Allen[3] and Hubbell[4] and the recent theoretical values of Scofield.

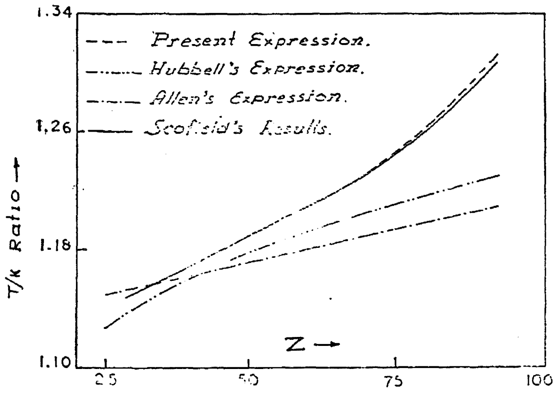

FIG. 1

It can be seen from the figure that the present trend differs from those of Allen and Hubbell in the high Z-region significantly. It can also be seen that the present values are in excellent agreement with the recent theoretical values of Scofield.

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1. Gordstein, G. W., *NBS Circular*, 1957, p. 583.
2. Davisson, C. M. and Evans, R. D., *Rev. Mod. Phys.*, 1952, 24, 79.
3. Allen, S. J. M., *Phys. Rev.*, 1926, 28, 907.
4. Hubbell, J. H., *NSRDS–NBS Report*, 1969, p. 29.
5. Rakavy, G. and Ron, A., *Phys. Rev.*, 1967, 159, 50.
6. Scofield, J. H., *UCRL Report* 51326, 1973.
7. Grigor'ev, E. P. and Zolotavin, A. V., *Soviet Phys. JETP*, 1959, 63 (9), 272.
8. Hultberg, S., *Arkiv fur Fysik*, 1959, 15, 315.
9. Radha Krishna Murty, V., *Ph.D. Thesis*, Andhra University, 1975.
10. Siva Sankara Rao, K., *Ph.D. Thesis*, Andhra University, 1975.

FRIEDEL-CRAFTS REACTION OF SELENIUM SULPHIDE

NUCLEOPHILLIC addition of sulphur into the organic moiety under Friedel-Crafts condition is known only in the case of benzene[1] while addition of selenium under similar conditions towards the synthesis of diaryl selenides has not been reported. The ditolyl and xylyl sulphides are synthesised by reduction of sulphoxides[2] or by the reaction of diorganomercury compounds with sulphur[3] or from the reaction of lead salts of organothio compounds with aryl halides[4]. The methods of introducing selenium into the organic moiety are summarised in literature[5,6]. It would be desirable to have the synthesis of both diaryl sulphide and selenide in one step and towards this end selenium sulphide as a starting material offers an advantage.

In the course of our studies on the synthesis and reactivity of selenium sulphides it has been found that octaatomic species, Se_xS_{8-x}, are inert towards many reactions though they possess interesting structures. The selenium sulphide, SeS_2, synthesised by published method[7], undergoes a variety of reactions and we wish to report here some of our results. The SeS_2 reacts with benzene, toluene and *p*-xylene in presence of anhydrous $AlCl_3$ to form diaryl sulphides in nearly quantitative amounts. No undesirable disulphide formation has been noticed. The course of the reaction can easily be monitored by the evolution of H_2S and quantitative precipitation of elemental selenium is observed at the completion of the reaction. The products are found to be pure from their bp/mp, elemental analysis, chromatographic and spectral methods. The reaction can be represented as :

$$RH + SeS_2 \xrightarrow{AlCl_3} R_2S + Se + H_2S.$$

No formation of diaryl selenides has been noticed though different reaction conditions such as varying mole proportions of the reactants, reaction time and temperature are employed.

In the case of toluene the GLC data showed that the product contained a mixture of three isomers. The major component (53%) has been characterised as the *para* substituted derivative since the infrared spectrum showed two absorptions at 1890 and 1780 cm^{-1} [8]. The minor components (35% and 12%) in the product, however, could not be identified by spectroscopic method nor the separation of the isomers was possible on account of their close boiling points. The minor components are recognised as di *ortho* and *ortho-para* substituted sulphides based on reactivity considerations.

The reaction seems to proceed through the formation of a reactive intermediate complex of the type,

$S_2^+[SeAlCl_3]^-$. This complex undergoes subsequent reactions as shown below:

$$S_2^+[SeAlCl_3]^- + RHAlCl_3 \rightarrow R{-}S{-}S{-}HAlCl_3 + AlCl_3 + Se$$

$$R{-}S{-}S{-}HAlCl_3 + RHAlCl_3 \rightarrow R{-}S{-}R + H_2S.$$

The composition of the complex gains support from the mass spectrum of SeS_2 which shows the predominant molecular ion being S_2^+. Further the postulation of such a complex also explains the nonformation of diaryl selenides owing to the poor nucleophilicity of selenium. The present work affords an elegant method towards the synthesis of simple diaryl sulphides in nearly quantitative yields.

Materials and Methods

All the chemicals used are of Analar grade and the solvents are distilled and dried before use. The infrared spectra are recorded on Perkin Elmer 237–B Infrared Grating Spectrometer. The GLC of diphenyl and tolyl sulphides are taken on 2–5% DC–11 silicone grease column at 150° and 174° respectively. The mass spectrum is recorded on MS 702 spectrometer using electron bombardment for molecular fragmentation. SeS_2 was synthesised[7] by the reaction of aqueous solutions of selenious acid with Na_2S at 5–7° and used without purification.

Diphenyl sulphide: About 20 g of anhydrous $AlCl_3$ was introduced into a three-necked flask containing 300 ml of benzene fitted with a stirrer, condenser and an addition flask containing 10 g SeS_2. The addition of SeS_2 was done slowly under an atmosphere of nitrogen and the reaction commenced immediately with the evolution of H_2S. The reaction was completed in about 2 hrs. The reaction mixture was hydrolysed with ice and the solvent removed under reduced pressure. This left behind a pale yellow liquid and it was passed through a column of silica gel to remove any possible disulphides. The product was distilled under vacuum and the pure colourless liquid distilling at 162–163°/18 mm was collected. Yield: 90%. Similar procedure was adopted for the synthesis of ditolyl, and dixylyl sulphides. Di *p*-tolyl sulphide was obtained as colourless liquid boiling at 179–180°/11 mm. Yield: 80%. Di *p*-xylyl sulphide was obtained as colourless crystals, m.p. 75–76°. Yield: 80%.

One of the authors (A. D.) is thankful to NCERT, New Delhi, for the financial assistance.

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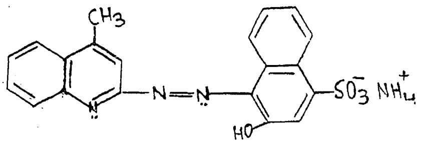
1. Blatt, A. H., *Organic Synthesis*, Collective Volume II, John Wiley, N.Y., 1950, p. 242.
2. Hampson, G. C., Farmer, R. H. and Sutton, L. E., *Proc. Roy. Soc. London*, 1933, 142 A, 147.
3. Ziegler, J. H., *Ber.*, 1895, 28, 1674.
4. (*a*) Bourgeois, *Ibid.*, 1895, 28, 2312; (*b*) Martinowicz, Z., *Chem. Zentr.*, 1910, 2, 1048.
5. Agenas, L. B., In *Organic Selenium Compounds; Their Chemistry and Biology*, Ed. by Klayman, D. L. and Gunther, W. H. H., John Wiley, N.Y., 1973, p. 173.
6. Kudchadker, M. V. and Irgolic, K. J., In *Selenium*, Ed. by Zingaro, R. A. and Cooper, W. C., Van Nostrand, Reinhold Pub. Co., N.Y., 1974, p. 408.
7. Krafte, M. Ya., Bordina, G. M. and Rubina, Z. O., *USSR*, Feb. 1, 1960, 125, 797.
8. Avram, M. and Mateescu, G. D., *Infrared Sprectroscopy, Applications in Organic Chemistry*, Wiley, Interscience, 1970, p. 217.

ANALYTICAL POTENTIALITIES OF A NEW HETEROCYCLIC AZO DYE: 1-(2-LEPIDYLAZO)-2-NAPHTHOL-4-AMMONIUM SULPHONATE (LANAS) AS A METALLOCHROMIC INDICATOR FOR TITRIMETRIC DETERMINATION OF MERCURY (II) WITH EDTA

HETEROCYCLIC azo dyes introduced recently[1] find intensive applications in analytical chemistry for micro determinations of metal ions spectrophotometrically[2] or complexometrically[3-5]. Only very few water soluble heterocyclic azo dyes have been explored so far as analytical reagents for micro determination of metal ions. In this communication, a new water soluble heterocyclic azo dye, 1-(2-lepidylazo)-2-naphthol-4-ammonium sulphonate (LANAS) has been synthesised, characterised and used as metallochromic indicator for complexometric determination of meercury (II).



(I)

TABLE I

Determination of mercury (II) with EDTA : Precision and accuracy

Sl. No.	Amount of Hg(II) in mg	Standard deviation Si	Variation coefficient	Relative width of confidence interval	% Relative error
1.	20	0·038	0·18	22·02 ± 0·015	+0·1
2.	100	0·106	1·06	100·1 ± 0·04	+0·1
3.	200	0·26	1·30	200·4 ± 0·11	+0·2
4.	4	0·006	0·15	4·008 ± 0·002	+0·2

LANAS (I) has been synthesised by condensing 2-hydrazino lepidine and sodium salt of 1, 2-naphthaquinone-4-sulphonic acid in presence of dilute hydrochloric acid and neutralizing the resulting solution with dil. ammonium hydroxide. The purity of LANAS has been checked by thin layer chromatography. The analysis for C, H, N (Found C = 59·43%, N = 13·51%, H = 4·5% ; calculated C = 58·53%, N = 13·66%, H = 4·39%) and I.R. spectrum (3650–3540 cm^{-1} bonded OH ; 1600 cm^{-1} —N=N— ; 1500 cm^{-1} $-CH_3$ rocking ; 1400 cm^{-1} NH_4^+ ; 1220, 1280 cm^{-1} $-SO_3^-$; 1000, 1050 cm^{-1} C–O) for LANAS justify the structure (I) for LANAS.

Mercury (II) forms water soluble blue coloured complex with LANAS. The blue colour of Hg(II)–LANAS is discharged quickly on adding EDTA solution. This has been taken advantage of in developing a sensitive and rapid method for *titrimetric* determination of mercury (II).

The pH and temparature ranges found suitable for an accurate determination are 6·0 to 7·5 (hexamine or pyridine–nitric acid buffer) and 0 to 70° C respectively. At higher temperatures the amount of EDTA used is less and below 0° C the end point is sluggish. Two to three drops of 0·01% of LANAS in distilled water are sufficient to cause a sharp colour change at the end point. The minimum and maximum amount of mercury that can be titrated accurately are 1 mg and 200 mg respectively.

Recommended Procedure

To aliquots containing mercury (II) add two drops of 0·01% LANAS solution and a few drops of dilute nitric acid till yellow colour is obtained. Then add sufficient amount of hexamine or pyridine-nitric acid buffer (former is preferred) till blue colour is obtained (pH 6·0 to 7·5). Titrate this with EDTA solution till the blue colour changes sharply to yellow. The titration precision is characterized by the standard deviation of single determination, Si, the variation coefficient %, Si·100/$\bar{x}$, the confidence interval and its width for a statistical significance of $\alpha = 0·005$. The titration accuracy is expressed by relative error $(\mu - \bar{x})$. $100/\mu$, where μ is the accurate value.

Results and Discussion

The following foreign ions did not cause any interference in the titration of 100 ppm of mercury (II), when they were present in amounts in ppm (in parenthesis) :

Bromide (1000), Fluoride and Sulphate (5000), Nitrite and Phosphate (2000), Acetate (large excess), Oxalate (5000), Tartrate (4500), Citrate (4000) and Sulphite (4000).

Sulphide, thiosulphate, iodide and cyanide interfere seriously. Hydroxyl amine (3 ml of 0·1 M), thiourea (2 ml of 0·1 M) do not interfere. Ascorbic acid gives permanent bluish turbidity.

Ruthenium, rhodium and osmium (150), palladium (20), platinum (500), tungsten (800), Mo(VI) and rare earths (5), alkaline earths (250) also do not interfere. Other metals interfere and the interference in a few cases has been overcome through masking. The amount of cations which have been masked are as follows :

Iron (III) (300)*, lead (II) (500)**, aluminium (III) (400)*, and bismuth (III) (100)***. Attempts to mask zinc and cadmium were unsuccessful.

Visual indicator reactions are not widely used for micro determination of mercury[6]. The present titrimetric method developed for mercury (II) is rapid and sensitive and many complexing anions do not interfere.

One of the authors (IS) is thankful to C.S.I.R., New Delhi, for providing necessary financial assistance.

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* Masked with fluoride.

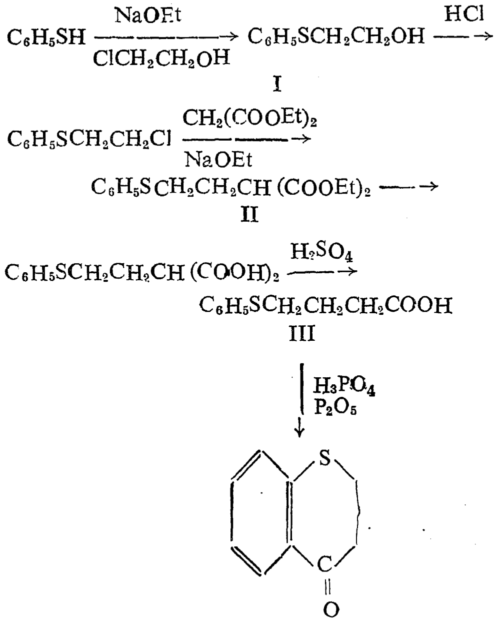
** Masked with sulphate or phosphate.

*** Masked with chloride.

1. Anderson, R. G. and Nickless, G., *Analyst*, 1967, 92, 1093.
2. Flaschka, H. A. and Barnard, A. J., *Chelates in Analytical Chemistry*, Marcel Dekker, Inc., New York, 1972, Vol. 4.
3. —, *EDTA Titrations*, Pergamon Press Ltd., Oxford, 1964.
4. Schwarzenbach, G., *Complexometric Titrations*, Methuen and Co. Ltd., London, Inc., New York, 1960.
5. Welcher, F. J.. *The Analytical Uses of Ethylenediamine Tetraacetic Acid*, D. Van Nostrand Co. Inc., Princeton, New Jersey, New York, 1961.
6. Chilov, S., *Talanta*, 1975, 22, 205.

SYNTHESIS OF 5-HOMOTHIOCHROMANONE

In connection with our work on the kinetics of acetylation of hydroxy sulphones, we required 5-homothiochromanone. The present work describes a new method for its synthesis and the scheme followed is given below.



$$C_6H_5SH \xrightarrow[ClCH_2CH_2OH]{NaOEt} C_6H_5SCH_2CH_2OH \xrightarrow{HCl}$$

I

$$C_6H_5SCH_2CH_2Cl \xrightarrow[NaOEt]{CH_2(COOEt)_2} C_6H_5SCH_2CH_2CH(COOEt)_2 \longrightarrow$$

II

$$C_6H_5SCH_2CH_2CH(COOH)_2 \xrightarrow{H_2SO_4} C_6H_5SCH_2CH_2CH_2COOH$$

III

$$\downarrow H_3PO_4,\ P_2O_5$$

γ-Phenylmercaptobutyric acid.—*β*-Hydroxyethyl phenyl sulphide (I) was obtained from thiophenol and ethylene chlorohydrin by adopting the procedure of Smith and Davis[1]. A mixture of I and concentrated hydrochloric acid was refluxed to give *β*-chloroethyl phenyl sulphide[1] (II); b.p. 123–124°/15 mm. Diethyl malonate (8 g) was added from a dropping funnel to sodium ethoxide in absolute alcohol (1·5 g of sodium and 50 ml of absolute alcohol). To this II (8 g) was added slowly and refluxed for an hour, cooled and after the addition of aqueous potassium hydroxide (25%, 100 ml), it was refluxed again for 4 hours and then neutralized with concentrated hydrochloric acid. The dicarboxylic acid separated as a solid; m.p. 156–158°. A mixture of the dicarboxylic acid (2·7 g), concentrated sulphuric acid (20 ml) and water (50 ml) was refluxed for 4 hours and on cooling γ-phenylmercaptobutyric acid (III) separated as a solid; m.p. 61–62° (Found: C, 61·1; H, 6·4%. $C_{10}H_{12}SO_2$ requires: C, 61·2; H, 6·2%). Cagniant and Deluzarche[2] obtained this acid by the hydrolysis of $C_6H_5S(CH_2)_3CN$; m.p. 63°.

5-Homothiochromanone.—A solution of III (6 g) in syrupy phosphoric acid (20 ml, *d* 1·750) was made by gentle heating and phosphorus pentoxide (50 g) gradually but quickly added with vigorous shaking. Large quantity of heat was evolved and a brown paste obtained. After 3½ hours, ice was added to the mixture. The product was isolated by extraction with ether; the ethereal solution was washed with aqueous sodium carbonate and then with water, dried and evaporated and 5-homothiochromanone was obtained as an oil; b.p. 180–181°/24 mm (yield 3·5 g). (Found: C, 67·6, H, 5·8%. $C_{10}H_{10}SO$ requires: C, 67·4, H, 5·7%). Its semicarbazone had m.p. 212–214°. Cagniant and Deluzarche[2] obtained 5-homothiochromanone from the acid chloride of (III) using aluminium chloride and reported b.p. 175–176°/22 mm; semicarbazone, m.p. 213°.

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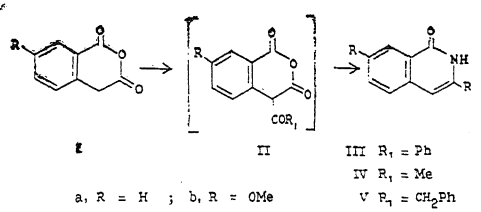
1. Smith, L. I. and Davis, Jr., H. R., *J. Org. Chem.*, 1950, 15, 824.
2. Cagniant, P. and Deluzarche, A., *Compt. Rend.*, 1946, 223, 677; *Chem. Abstr.*, 1947, 41, 1669.

A NEW METHOD FOR SYNTHESIS OF 3-PHENYLISOQUINOLONES

3-Phenylisoquinolone (III*a*) assumes importance because of anti-inflammatory, antibacterial, antiprotozoal and antifungal activity reported[1] recently for its various N-substituted derivatives that can be synthesised[1] directly from III*a*. Synthesis of III*a* was first reported by Gabriel[2] by action of ammonia on 3-phenylisocoumarin but because of low yield of the latter by the methods[8,9] employed then, it did not receive much acceptance and that led to development of many other methods[1-7] for synthesis of III*a*. A better synthesis of 3-phenylisocoumarin by interaction of I*a* with benzoic anhydride in presence of triethylamine was recently reported[10] from this laboratory and using it as modification, Gabrial's synthesis of III*a* wiil be much superior to other syntheses including the one patented[1] recently both from the point of steps involved and the starting material used.

We now report a more direct synthesis of III*a* in one step from I*a* in a yield of about 70%, much superior to that even of 3-phenylisocoumarin[10] from I*a* and the reaction is very clean, with easy work up and free from tarry products. It consists of adding benzoyl chloride (5·2 ml) in dry chloroform (16 ml) slowly to a mixture of I*a* (4 g) and pyridine (12 ml) at 0°, stirred mechanically. After stirring it under this condition for an hour it is to be treated with liquor ammonia (100 ml), then to be refluxed for 2 hours and kept at room temperature overnight, when 3-phenylisoquinolone (III*a*) separates out (washed with dil. HCl to remove pyridine and crystallised from ethanol as needles, yield 3·6 g, m.p. 199–200. Lit.[2] m.p. same. Found: C, 81·1; H, 5·3 and N, 6·3. $C_{15}H_{11}ON$ requires: C, 81·4; H, 5·0 and N, 6·3%). The reaction is taking place by initial benzoylation[10] of the reactive —CH_2 of I*a* to give 4-benzoylisochroman-1, 3-dione (II*a*, R_1 = Ph) which reacts with ammonia *in situ* with decarboxylation by the same mechanism as reported[11] for the transformation of 4-acetylisochroman-1, 3-dione to 3-methylisoquinolone. The reaction is of general applicability and with the appropriately substituted homophthalic acids and different acid chlorides, desired 3-phenylisoquinolones with substituents in benzene part and 3-phenyl nucleus can be directly synthesised in one step.



The reaction has been extended to 4-methoxyhomophthalic anhydride (I*b*) with equal success to give, in very good yield, 7-methoxy-3-phenyl-1 (2H)-isoquinolone (III*a*) that has not been described so far in lit. (crystallised from benzene as needles, m.p. 206–07°; Found: C, 76·2; H, 5·0 and N 5·3. $C_{16}H_{13}O_2N$ requires: C, 76·4; H, 5·2 and N, 5·5%). 3-Methylisoquinolone (IV*a*) and its 7-methoxy derivative (IV*b*) whose syntheses were reported[11] earlier from I*a* and I*b* respectively *via* 4-acetylisochroman-diones can be obtained in excellent yield, in one step directly from I*a* and I*b* respectively by the above procedure using acetyl chloride. In the same way, 3-benzylisoquinolone[12] (V*a*) and its 7-methoxy derivative[13] (V*b*) (m.p. 220–22°) whose syntheses from I*a* and I*b* respectively were reported *via* 3-benzylisocoumarins can now be obtained in excellent yield from I*a* and I*b* respectively in one step by the above procedure using phenylacetyl chloride.

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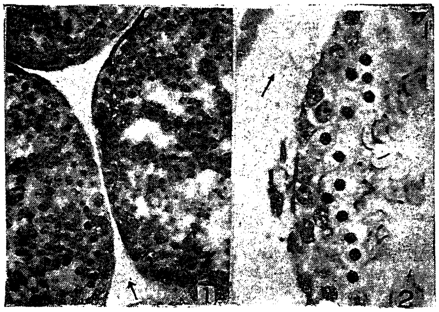
1. Coyne, W. E. and Curie, J. W. (Searle G. D. & Co. U.S. 3, 600, 394 Cl–260–288, CO 7*d*); *Chem. Abstr.*, 1971, 75, 118241 *f*.
2. Gabriel, S., *Ber.*, 1885, 18, 3471.
3. Ulrich, A., *Ibid.*, 1904, 37, 1692.
4. Wolbing, H., *Ibid.*, 1905, 38, 3853.
5. Wilson, J. W., Anderson, E. L. and Ullyot, G. E., *J. Org. Chem.*, 1951, 16, 800.
6. Boyce, W. T. and Lavine, R., *Ibid.*, 1966, 31, 3807.
7. Bogenloft, C., Kranberg, L. and Bengt, D., *Acta Pharm. Succia.*, 1969, 6, 489; *Chem. Abstr.*, 1970, 72, 11916 *r*.
8. Gabriel, S., *Ber.*, 1885, 18, 2445.
9. Graebe, C. and Trumphy, F., *Ibid.*, 1898, 31, 375.
10. Tirodkar, R. B. and Usgaonkar, R. N., *Curr. Sci.*, 1972, 41, 701.
11. — and —, *Indian J. Chem.*, 1972, 10, 1060
12. Belgaonkar, V. H. and Usgaonkar, R. N., *Ibid.*, 1975, 13, 336.
13. —, *M.Sc. Thesis*, Bombay University, 1973.

ABSENCE OF LEYDIG CELLS IN THE MATURE TESTIS OF THE LIZARD *LYGOSOMA HIMALAYANUM*

VARIOUS studies on interstitial cells of oviparous lizards from Kashmir have been made[1,2]. A similar study was undertaken on high altitude ovoviviparous lizards in order to draw some generalisations regarding the Leydig cell activity in the testicular cycle in lizards. About 120 specimens of *L. himalayanum* were examined during two successive years of 1972 and 1973.

The investigations into the testicular elements of *L. himalayanum* have apparently revealed a complete absence of the Leydig cells in the testes examined histologically in any month of the observation. They are completely wanting in their characteristic sites; the intratubular spaces (Figs. 1 and 2) and the subtunic areas. In the intertubular spaces a few fibrocytes were, however, noticeable, A complete absence of interstitial cells in *Sceloporus undulatus* has been reported[3] and the present observations lend a support to the generalisation that the interstitial cells are variable of all the testicular elements. Leydig cells may thus be absent in *L. himalayanum, S. undulatus* or rarely occurring in *Sceloporus occidentalis*[4].



FIGS. 1–2. Cross section testis (fixative, Smith's fluid and Stain, Mallory's tripple). Note the absence of Leydig cells (arrow), × 225 for Fig. 1 and × 500 for Fig. 2.

There has been a conjecture that the interstitial cells may be functionally connected with the puberty or attainment of sexual maturity, but in view of their absence in some lizards mentioned above, this type of functional relationship may not be attributable to interstitial cells alone. There is thus a lurking possiblity that in such forms where the interstitial cells are absent, some other testicular element may be responsible for the androgenic functions of the testes or even maintenance of puberty. This aspect is currently under investigation.

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1. Duda, P. L. and Opender Koul., *Jour. Herp.*, 1976, 10 (3), 181.
2. — and —, *Br. Jour. Herp.*, 1975 (In press).
3. Altland, P. D., Elisha, *J. Mitchell. Sci. Soc.*, 1941, 157, 73.
4. Wilhoft, D. C. and Quay, W. B., *J. Morph.*, 1961, 108, 95.

ON THE LOCALISATION OF HYDROXYSTEROID DEHYDROGENASES IN THE TESTIS OF THE SKINK, *MABUYA CARINATA*

$\Delta^{5}3\beta$- AND 17β-hydroxysteroid dehydrogenases are important enzymes in steroid metabolic pathway. Histochemical demonstration of these enzymes in the testis of many reptiles has implicated interstitial cells to be the principal sites of steroid biosynthesis[1-5]. However, there also exist reports which record these enzymes in the seminiferous tubules[6,7]. A recent report indicates the localisation of these hydroxysteroid dehydrogenases (HSDHs) in the interstitial cells of the testis in *Mabuya carinata*[8]. Since our earlier observations had revealed contradictory results[9], we undertook a detailed histochemical analysis of the steroidogenic site in the testis of the skink, *Mabuya carinata,* taking into consideration the reproductive cycle of the animal about which the previous report[8] does not mention. The present paper reports the distribution of lipids, $\Delta^{5}3\beta$-, 17β-HSDHs, G–6–PDH and $NADH_2$– diaphorase in the skink testis.

Frozen cryostat sections of the testis from mature skinks collected during different months of the year were incubated for HSDHs according to Baillie *et al.*[10]. The incubation medium consisted of the steroid substrate (5 mg/ml dissolved in dimethyl formamide), NAD (2 mg/ml) and NBT (1 mg/ml) in Tris buffer (PH 7·4). The substrates used for HSDHs were DHA, Pregnenolone, 17 α-hydroxy Pregnenolone (for $\Delta^{5}3\beta$-HSDH), Eatradiol-17β- and Testosterone (for 17β-HSDH). G-6-PDH and diaphorase were localised as given by Chayen *et al.*[11]; Sudan Black B method was employed for lipids. Suitable controls were used in each case.

Both $\Delta^{5}3\beta$- and 17β-HSDHs were distributed mainly in the seminiferous epithelium with all the substrates used while the interstitium hardly gave any positive reaction as could be judged from the formazan deposition (Fig. 1). However, G-6-PDH,

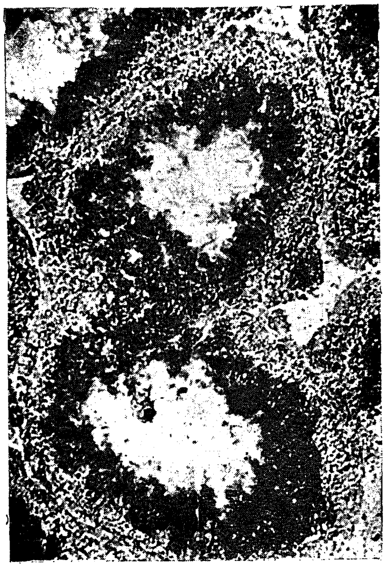

FIG. 1. T.S. of the skink testis showing $\Delta^{5}3\beta$-HSDH activity with Pregnenolone as the substrate. Note the activity confined mainly to the seminiferous epithelium, × 1,300.

diaphorase and lipids were mainly interstitial in their distribution though slight activity could be seen in tubular epithelium. While the distribution of G-6-PDH and diaphorase is in agreement with that of the previous report, the tubular localization of HSDHs contradicts their observation[8]. The occurrence of $\Delta^{5}3\beta$- and 17β-HSDHs in the seminiferous epithelium indicates the oxidative conversions of $\Delta^{5}3\beta$-hydroxy steroids to $\Delta^{4}3$keto forms, a basic step in steroid biosynthesis and interconversions of various estrogenic and androgenic steroids respectively. Our observations in *Mabuya carinata* seem to indicate seminiferous tubules as another possible site of steroid metabolism in this species and this is interesting in the light of previous reports which stress the importance of seminiferous tubules and their involvement in steroid biosynthesis[6,7,12].

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1. Callard, I. P., *J. Endocrinol.*, 1967, 37, 105.
2. Nandi, J., *Am. Zool.*, 1967, 7, 105.
3. Currie, C. and Taylor, H. L., *J. Morph.*, 1970, 132, 101.
4. Erpino, M. J., *Gen. Comp. Endocrinol.*, 1971, 17, 563.
5. Gouder, B. Y. M. and Nadkarni, V. B., *Indian J. Exp. Biol.*, 1974, 12, 326.
6. Simpson, T. H. and Wardle, C. S., *J. Mar. Biol. Ass. U.K.*, 1967, 47, 699.
7. Lofts, B., *Gen. Comp. Endocrinol. Suppl.*, 1972, 3, 636.
8. Gouder, B. Y. M. and Nadkarni, V. B., *Curr. Sci.*, 1976, p. 102.
9. Shivanandappa, T. and Sarkar, H. B. D., *All India Symp. Comp. Endocrinol, Waltair, Abstract*, 1975.
10. Baillie, A. H., Ferguson, M. M. and Hart, D. McK., *Developments in Steroid Histochemistry*, Academic Press, London, 1966.
11. Chayen, J., Bitensky, L. and Butcher, R. G., *Practical Histochemistry*, John Wiley & Sons, London, 1973.
12. Lofts, B. and Bern, H. A., In: *Steroids in Nonmammalian Vertebrates*, Ed. by Idler, D. R., Academic Press, New York, 1972.

ON THE NATURE OF THE CYST OF *LERNAEENICUS HEMIRHAMPHI* KIRTISINGHE

EVEN though a cyst and sheath are characteristic around the head and neck of lernaeid copepods[1-3], the exact nature and the biochemical composition of these structures are still unknown.

The head and neck of *Lernaeenicus hemirhamphi* are both buried within the body of the host, *Hemirhamphus xanthopterus* Val. The rest of the body comprising the genital region with the egg tubes and abdomen hang free in the sea water. When exposed *in situ* the head and neck of the parasite are seen embedded in thick, three-layered, fibrous, granular, connective tissue cyst formed by a pathological transformation of the host's body, probably resulting from tissue reaction caused by the irritation of the parasite (Fig. 1). The outer and middle layers are formed of fibro-elastic connective tissue composed of cells with long cytoplasmic processes and large and oval nuclei containing diffuse chromatin material. The innermost layer is formed of closely arranged collagenous fibres stained by eosin and acid dyes bearing nuclei placed far apart.

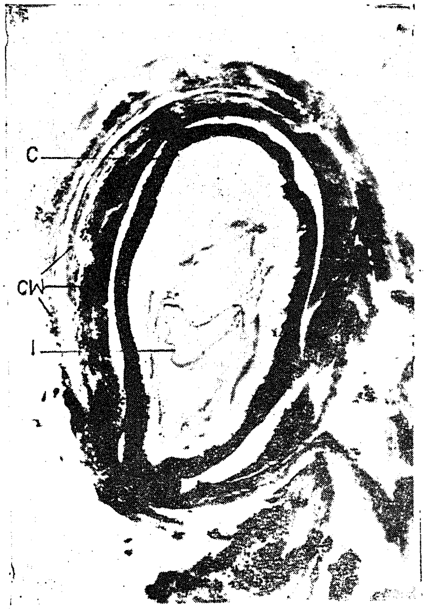

FIG. 1. T.S. passing through the neck of *L. hemirhamphi* showing the nature of the cyst, × 140. C, cuticle; CW, cyst wall; *i*, intestine.

Histochemical tests on the cyst from parasites which were embedded in the muscles of the fish (Table I) with the aid of periodic acid Schiff (PAS) technique for glycogen, mercuric bromphenol blue (MBM) for proteins, and Sudan black B (SB) for lipids showed that the outer and middle layers of the cyst wall are faintly PAS positive showing the presence of traces of glycogen. These layers further showed positive reaction with mercuric bromphenol blue indicating the presence of protein but did not give a consistent result for lipids with Sudan black B.

TABLE I

	PAS	MBM	SB
			—

+ Weak reaction ; ++ Moderate reaction ; ? = Doubtful reaction ; — Negative reaction.

The inner layer is moderately PAS positive and diastase resistant which suggest that this layer may consist of PAS positive inclusions such as glycolipids, mucoproteins or glycoproteins[4]. Mercuric bromphenol blue test shows sure indications of mucoproteins and glycoproteins in this layer.

Thus the indications are that the cyst of L.

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PHAEOISARIOPSIS GRISEOLA (SACC.) FERRARIS ON FRENCH BEANS

IN October 1975 French beans, *Phaseolus vulgaris* Linn. var. 'Contender' were grown in the University of Agricultural Sciences, Hebbal Campus, Bangalore. The maximum temperature of 28° C, minimum 18° C and R.H. 60–90% were recorded during that period. There was 61·5 mm rainfall and about 13% soil moisture.

An interesting dematiaceous hyphomycete was collected on leaves and stems of *P. vulgaris* which on examination was found to be *Phaeoisariopsis griseola* (Sacc.) Ferraris. The genus *Phaeoisariopsis* was erected by Teodoro Ferraris in 1909 based on *Isariopsis griseola* Sacc. He has transferred three more species namely, *grayana* Ell., *mexicana* Ell. and Ev. and *pilosa* Earle. Srinivasan[2] recorded *I. griseola* on *P. vulgaris* from Tamil Nadu, which is a *nomen dubium* and insufficiently described. He has further reported that varieties 'Black valentine', 'Bountiful yellow' and local french beans were susceptible to the disease. Hocking[1] described a new virulent form causing circular leaf-spots of french beans, commonly found on leaves and petioles from Tanzania. A short description of the fungus is given below.

Phaeoisariopsis griseola (Sacc.) Ferraris (Fig. 1) in *Ann. Mycol.*, 1909, 7, 280; Ellis, M. B., *Dematiaceous Hyphomycetes*, 1971, p. 268.

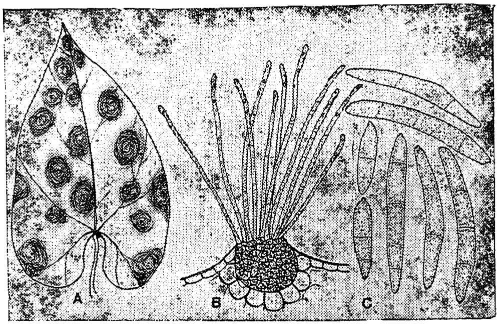

FIG. 1. *Phaeoisariopsis griseola* (Sacc.) Ferraris. A, Habit; B, Conidiophores emerging from the stroma (× 150); C, Conidia (× 330).

Foliicolous and caulicolous. Spots numerous, amphigenous, irregularly circular, 5–20 mm long and 5–15 mm wide, coalesced to form irregular patches involving complete lamina. Fructifications mostly hyphophyllous, a few epiphyllous. The spots appear mostly on the first leaves in the beginning and later spread to other leaves. The spots appear water-soaked under humid conditions. In advanced stage of infection, most of the leaves and petioles are affected resulting in complete defoliation. As a result the yield is reduced up to 80% and the plants die prematurely.

Colonies effuse, brown and hairy. Mycelium immersed. Stroma present, prosenchymatous and brown. Setae and hyphopodia absent. Conidiophores macronematous, mononematous, forming synnemata up to 520 mμ long, individual threads unbranched, straight or flexuous, pale to mid-brown, smooth, about 3·75 mμ thick near the base, swelling up to 5·75 mμ near the apex, 2–4 septate. Conidiogenous cells polyblastic, integrated, terminal, sympodial, cylindrical to clavate, cicatrized, scars flattened against the side of the conidiogenous cells.

Conidia solitary, dry, acropleurogenous, simple mostly obclavate, conico-truncate at the base, pale olive to olivaceous brown, smooth, 1–5 septate, 30–60 (55) × 6·5–7·0 (6·5) mμ in dimension.

On living leaves and stems of *P. vulgaris*. October 10, 1975. UAS, Hebbal Campus, Bangalore. Leg. P. C. Hiremath. Specimens deposited in Herbaria of the Commonwealth Mycological Institute, Kew and in the Department of Plant Pathology as IMI 198914 and MYSP 1961 respectively.

Further studies regarding host-range, chemical control and other aspects are in progress.

Authors are grateful to Dr. M. B. Ellis, Commonwealth Mycological Institute, Kew, for confirming the identification of the pathogen and to Dr. H. C. Govindu, Head, Department of Plant Pathology, for encouragement.

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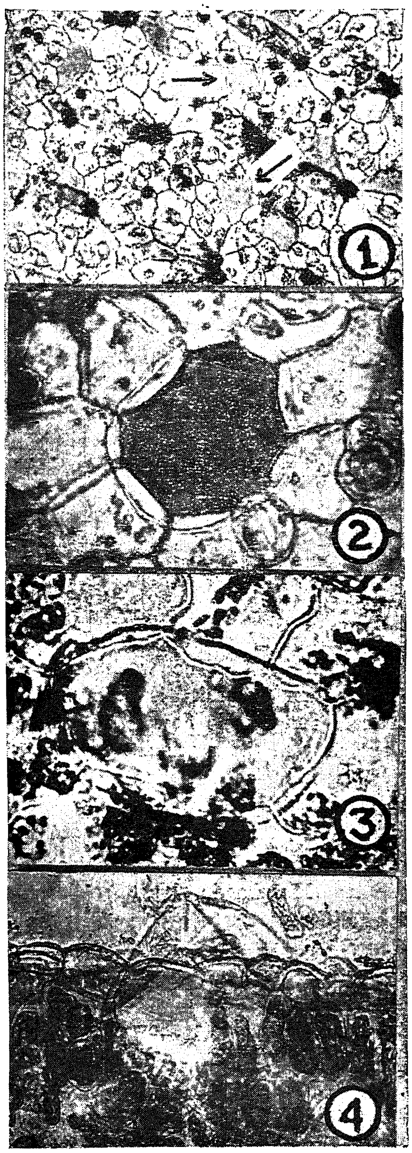
1. Hocking, D., *Pl. Dis. Reptr.*, 1967, 51 (4), 276.
2. Srinivasan, K. V., *Curr. Sci.*, 1953, 22 (1), 20.

UNICELLULAR STORAGE IDIOBLASTS NORMAL AND MUTAGEN INDUCED *CROTALARIA* L. SPECIES

THE objective of this communication is to report the occurrence of unicellular storage idioblasts in foliar epidermis of normal and mutagen induced *Crotalaria* species. With the main purpose of studying the comparative efficiency of physical and chemical mutagens on *Crotalaria* L. species, seeds of *C. juncea* L. cv. PLC 31, *C. medicaginea* L. var. 1. *luxuriance*, 2. *neglecta* and 3. *typica*, and *C. triquetra* Daltz. were treated with gamma rays (γ-rays; 20–100 kR) and dimethyl sulphate (DMS; 0·2–0·4%), and plants were raised in 12″ earthenware pots. The treatment produced varying degrees of lethality and morphological variability including phyllody where the leaves were compactly arranged and very much reduced in size, and the floral parts were proliferately modified into leaf-like structures in M_I generation. In the epidermal studies of these induced variants and normal plants, storage idioblasts were observed in control and treated plants of *C. medicaginea* var. *typica*, and γ-rays (80 and 100 kR) and DMS (0·4%) induced phyllody types of *C. juncea* (Figs. 1–4) with a comparatively high frequency on the adaxial surface of the leaves.

The idioblasts are generally larger than the tabular epidermal cells and more or less spherical in shape. Their upper surfaces protrude slightly above the epidermal cells (Figs. 2, 3) and their bases are surrounded by palisade parenchyma (Fig. 4). The cell wall is comparatively thick and the cytoplasm shows greater chromaticity in contrast to that of surrounding epidermal cells. It is presumed that the contents of these cells may impart the characteristic violet pigmentation to the leaves.



FIGS. 1–4. Fig. 1. Surface view of storage idioblast cells in upper foliar epidermis of *C. medicagenea* var. *typica*. × 150, Fig. 2. Single idioblast magnified, × 500. Fig. 3. Same, in gamma rays induced phyllody type of *C. juncea*, × 500. Fig. 4. Same, in side view from transection of leaf, × 500.

Formation of epidermal idioblasts has been considered to be related with micro-environmental influences[1]. The concept seems plausible, since in untreated populations, they are present only in *C. medicaginea* var. *typica*, a prostrate from having its own micro-environmental surroundings different from those of the erect forms. Their formation in induced phyllody types of *C. juncea* is caused by morphological changes brought about by lethal doses of mutagen treatment. It results in destruction of biological activity of growth regulators[2,3] and induction of physiological and biochemical disturbances[4]. Hawever, the change is not genetical but morphologically epigenetic since, induced phyllody types are completely sterile without fruit and seed setting.

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Jaipur, *June* 10, 1976.

---

1. Mitchell, R. S., *Amer. J. Bot.*, 1971, 58, 342.
2. Skoog, F., *Science*, 1934, 79, 256.
3. Sideris, E. G., Kleinhofs, A. and Nilan, R. A., *Radiat. Bot.*, 1969, 9, 345.
4. Gunckel, J. E. and Sparrow, A. H., *Brookhaven Symp. Biol.*, 1954, 6, 252.

MODIFICATION OF SEX EXPRESSION BY KINETIN, MORPHACTIN AND UREA IN CASTOR (*RICINUS COMMUNIS* L.)

CHEMICAL control of sex expression was achieved mostly in cucurbits[1-3]. The work on monoecious plants other than cucurbits is scanty. In castor, separate male and female flowers are borne on the same plant so that the sexes are separated flower for flower but not plant for plant. While investigating the physiological basis of sex expression, it was intended to modify the sex ratio by growth regulators.

Castor seeds var. Aruna were selected for the investigation. Morphactin, kinetin, and urea were sprayed at different stages of vegetative growth. As castor is a 90 days crop, the total vegetative period was divided into six stages of 15 days interval, the fifth stage being the flower initiation stage. In general the untreated castor plants showed a ratio of 4 : 1 (Male : Female) flowers. Kinetin (10 ppm) reduced the maleness significantly although femaleness was not increased with a ratio of 2 : 1 (M : F). Urea (2–3%) also modified the sex ratio to nearly 2 : 1, similar to that of kinetin. In monoecious plants kinetin may not increase femaleness but reduces male tendency. It is also evident that nitrogen in any form (urea and kinetin) may reduce the maleness in monoecious plants. Earlier work by Gopala Rao[4] indicated that decapitation of shoot apex in castor increased the male tendency possibly by reducing the auxin level and increasing the gibberellin level.

An interesting finding is that morphactin at high concentrations (500 ppm), when sprayed only once just before flowering, reduced femaleness giving a ratio of 10 : 1 (M : F) and when sprayed throughout vegetative stage at different intervals of 15 days, during the 90 days period, also reduced the femaleness giving a ratio of 10 : 2 (M : F) at a concentration of 50 ppm. In general, it was found that treatment with kinetin, morphactin or urea delayed anthesis period. It is finally concluded that nitrogen supply to monoecious plants reduces maleness and higher concentration of morphactin (500 ppm) sprayed once, prior to flower initiation, reduces femaleness drastically.

•The authore rre thankful to Prof. V. S. Rama Das for his encouragement and for facilities.

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Tirupati, *March* 5, 1976.

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1. Rudich, J., Halvy, A. H. and Kedar, N., *Planta*, 1969, 86, 69.
2. Peterson, C. E. and Anhder, L. D., *Science*, 1960, 131, 1973.
3. Holevy, A. H. and Rudich, Y., *Physiol Plant.*, 1967, 20, 1052.
4. Gopala Rao, P., *Science and Culture*, 1969, 35 (7), 326.

TRYPANOSOMA ACANTHOBRAMAE N. SP. FROM A FRESHWATER FISH, *ACANTHOBRAMA MARMID* HECKEL (FAMILY: CYPRINIDAE) FROM THE RIVER TIGRIS, IRAQ

TRYPANOSOMES have been reported from freshwater and saltwater fishes in various parts of the world. During a survey of fish parasites in 1973–74, the authors came across a new species of *Trypanosoma* from the blood of a Cyprinid fish, *Acanthobrama marmid* Heckel.

Materials and Methods

For the study of trypanosomes, thin blood films were made on clean glass slides from the blood obtained by clipping the caudal peduncle of the fish. The films were air-dried, fixed in methyl alcohol and stained with Giemsa's stain. Camera lucida drawings were made as well as phase-contrast photographs. ,

Observations

The organisms are slender and attenuated at both the ends. Their configuration varies, some of them have a "C"-shaped body, while others are "S"-shaped. No polymorphism is observed and no dividing stages found in the blood smears.

The average measurements of the methanol fixed specimens are :

Body length, 21·71 to 30·06 (average 25·89) microns ; the length of the free flagellum, 18·37 to 21·71 (20·37) microns ; total length from tip to tip of the organism, 33·4 to 50·1 (43·0) microns ; the length of the nucleus, 3·34 to 5·85 (5·01) microns and its width, 1·67 to 2·51 (2·17) microns ; the distance from nucleus to kinetoplast, 6·16 to 9·24 (7·60) microns and the nuclear index, 0·75 to 1·13 (0·87).

The nucleus is sausage-shaped, stains light purple in Giemsa's stain and usually occupies the entire width of the body. It is normally located near the inner border of body curvature.

The cytoplasm is vacuolated and stains blue, the vacuolations vary. In some, large vacuoles are present near the posterior border of the nucleus, while in others they are small. No myonemes were observed. Numerous reddish-purple granules considered to be chromatic granules are present between the nucleus and the kinetoplast.

The kinetoplast is usually rounded or ovoid, sometimes conical, staining dark red. It measures about 1·68 microns in length. The flagellum originates from the kinetoplast and trails anteriorly, bordering the undulating membrane and extending beyond the body as a free structure. The undulating membrane stains light red and is clearly outlined.

The infection is more pronounced in the summer months.

Discussion

Since the discovery of *Trypanosoma* in the blood of the trout (*Salmo faris*), a large number of trypanosomes have been seen in freshwater and saltwater fishes in various parts of the world.

The general morphology of these is the same. They have usually long and narrow bodies, and vary greatly in size. The trypanosome of the pike, *T. remaki* measures only 15 microns, while that of the ray about 130 microns, but the majority of species have a length of about 50 microns.

Description of trypanosomes of North American freshwater fishes[1] makes no mention of their occurrence in the Cyprinidae. Another report on diseases of fishes[2] mentions the occurrence of *T. danilewskyi* in the carp, *Cyprinus carpio* and *T. phoxini* in the minnow, *Phoxinus phoxinus*, but none from *Acanthobrama marmid*. Only one distinct species, *T. occidentalis*, has been described in North America[3], from two species of sculpins, *Cottus gulosus* and *C. rhotheus*, and one from stickleback, *Gastrosteus aculeatus*, from the Pacific coast. *Trypanosoma remaki* was also reported from *Esox lucius*, *E. reticulatus* and probably other species[4]. Another species, *T. percae*, described in *Perca fluviatilis* and from the brook trout[5], resembles to *T. rotatorium* of the frog. More recently, *T. catostomi* has been reported from the white sucker, *Catostomus c. commersoni*[6].

The trypanosome reported here from *Acanthobrama marmid* has some resemblance in the body length to *T. danilewskyi*.

Till now, so far as the authors are aware, no trypanosome has been described from *Acanthobrama marmid*, and none, at any rate, from Iraq or neighbouring countries[7]. Also, reports on the parasites of freshwater fishes[8-9] make no mention of any trypanosomes from any of them.

The trypanosome from *A. marmid* is, therefore, considered as a new species and designated as *Trypanosoma acanthobramae*.

Locality : River Tigris, Mosul (Iraq).

Type specimens : Syntypes deposited with Parasitology Section, Department of Biology, Mosul University, Mosul, Iraq.

Department of Biology, University of Mosul, Mosul, Iraq, *March* 6, 1976.
AKHLAQ A. WARSI.
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---

1. Hoffman, G. L., *Parasites of North American Freshwater Fishes*, University of California Press, Berkeley, 1970, p. 486.
2. Van Dujin, *Diseases of Fishes*, Iliffe Book Ltd., Derset House, Stamford Street, London, S.E. 1.
3. Becker, C. D., *J. Protozool.*, 1967, 14, 153.
4. Kudo, R. R., *Protozoology*, 5th Ed., Charles C. Thomas, Springfield, Illinois, 1966.
5. Fantham, H. B., Porter, A. and Richardson, L. R., *Parasit.*, 1942, 34, 199.
6. Daly, J. J. and Degiusti, D. L., *J. Protozool.*, 1971, 18, 414.
7. Becker, C. D. Personal Communication, 1975.
8. Herzog, P. H., *Arch. Fischereiwiss.*, 1969, 20, 132.
9. Shamsuddin, M., Nader, I. A. and Al-Azzawi, M. J., *Bull. Biol. Res. Centre*, Baghdad, 1971, 5, 66.

CHINA DODDER (*CUSCUTA CHINENSIS* LAMK.) ON COWPEA—A NEW RECORD

IN a survey of the vegetation in and around the Dairy Farm area of Visakhapatnam, A.P., a very severe attack by China dodder of a field of cowpea—an important pulse, fodder and green manure crop[1] (*Vigna sinensis* Endl.)—has been met with. This is a recent addition to the Flora of Visakhapatnam, a new record for Andhra Pradesh and a new record of cowpea acting as a host to this dodder. The parasitised plants show wrinkled and yellowish leaves, branches and pods and die prematurely. Pods show stunted growth and dwarfing. The parasite flowers profusely (Fig. A). This first appeared here in 1974 introduced along with some forage crops like *Melilotus* and *Medicago*. Data about nomenclature and description were provided by Santapau[2].

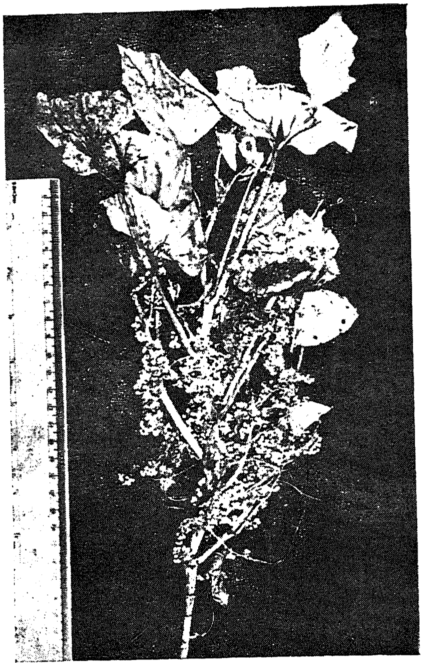

FIG. A. *Cuscuta chinensis* Lamk. (China dodder) on cowpea.

One of us (V. S. Raju) is grateful to the CSIR for the award of a fellowship.

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---

1. Krishnaswamy, N., In *Pulse Crops of India* (Ed. P. Kachroo), ICAR, 1970, p. 201.
2. Santapau, H., *J. Bombay Natural Hist Soc.*, 1947, 47, 337.

KARYOLOGICAL STUDIES IN *PANCRATIUM LONGIFLORUM* L.

Pancratium longiflorum L., a subtemperate and tropical member of Amaryllidaceae, is a dwarf bulbous plant of medicinal avail. It bears linear leaves and white scented flowers. Of the fourteen species of this genus, cytological work is confined to *P. illyricum*[2,3], *P. zeylanicum*[4], *P. triflorum*[6] and an undetermined species[5]. Since *P. longiflorum* L., as far as the authors are aware, has not been cytologically explored, the present investigation is undertaken and its karyomorphology is detailed in this communication.

From the observation of somatic metaphase preparations, it was observed that the chromosome number of the current species is $2n = 22$ (Fig. 1) and the karyotype comprises the following eleven pairs of chromosomes (Fig. 2).

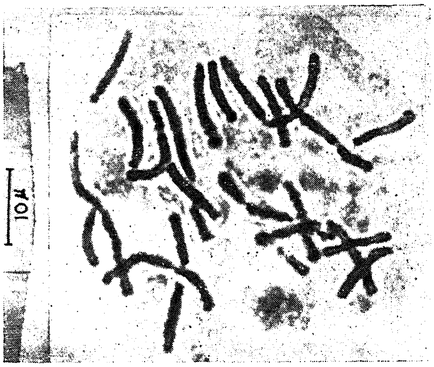

FIG. 1

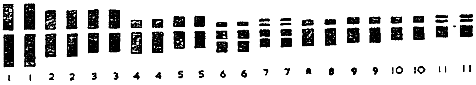

FIG. 2

FIGS. 1–2. Fig. 1. Mitotic complement of *Pancratium longiflorum showing* $2n = 22$ chromosomes, × 720. Fig. 2. Idiogram of *Pancratium longiflorum*.

(1) Two pairs of long chromosomes (11·2 μ and 8·8 μ) having median primary constrictions; (2) one pair of medium chromosomes (8·0 μ) with median centromeres; (3) three pairs of medium sized chromosomes (6·4 μ, 4·8 μ and 4·4 μ) having nearly subterminal primary constrictions; (4) two pairs of medium sized chromosomes (6·0 μ and 4·4 μ) having submedian primary constrictions; (5) one pair of medium sized chromosomes (5·6 μ) having almost subterminal primary constrictions and secondary constrictions on the long arms; (6) one pair of medium sized chromosomes (5·2 μ) having subterminal primary constrictions and each with two secondary constrictions, one on the long arm and the other on the short arm; (7) one pair of short chromosomes (4·0 μ) with submedian primary constrictions and secondary constrictions on the short arms.

Of the eleven pairs. two pairs are long, eight are medium and one is short. Among them, three pairs have median constrictions, three possess submedian ones and five pairs subterminal ones (3 M + 3 Sm + 5 St), thus displaying a symmetrical karyotype.

As in other taxa of Amaryllidaceae, polymorphic races are also evident in the genus *Pancratium*. In *P. illyricum*, the diploid ($2n = 22$) and the tetraploid ($2n = 44$) races were reported by Brumfield and Sato respectively. A resembling variation has been recorded by Venkatakrishnan in the somatic chromosome number, that is $2n = 44$ in Shornur clone and $2n = 48$ and 56 in Malapuram clones of *Pancratium triflorum*. However, in the present species (*P. longiflorum*) no such deviation in chromosome number has been observed by the authors. Sharma and Ghosh (1954) studied an undetermined species of *Pancratium* and reported 48 somatic chromosomes of which six are long, twenty-eight medium and fourteen short. Sharma and Bal (1956) reported $2n = 22$ in *P. zeylanicum* and observed two pairs of long, five pairs of medium and four pairs of short chromosomes. From the present study it becomes obvious that the chromosome number in *P. longiflorum* is in agreement with that of *P. zeylanticum*[4] and *P. illyricum*[2] but for the eight pairs of medium chromosomes and one pair of short chromosomes. The present record of three pairs of chromosomes with secondary constrictions of which a pair having secondary constrictions both on long and short arms in *Pancratium longiflorum* should be reckoned as the first for the genus *Pancratium*.

The basic chromosome number in the allied genera *Hymenocallis, Crinum* and *Eucharis* (subtribe Cyathiferae of Bentham and Hooker[1]) is 11. From the available information it appears, as though, the lowest chromosome number for *Pancratium* is $2n = 22$ (Brumfield, 1941 ; Sharma and Bal, 1956 ; present report). Thus from the chromosome report and also from the occurrence of eleven types of chromosomes in the karyotype it seems reasonable to deliberate that 11 is the basic number for the genus *Pancratium*.

One of the authors (N. Lakshmi) is grateful to the Indian Council of Agricultural Research, New Delhi, for financial assistance during the course of this investigation.

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Waltair, *February* 24, 1976.

---

1. Bentham, G. and Hooker, J. D., *Genera Plantarum*, London, III, 1883, p. 671.
2. Brumfield, R. T., *Amer. Journ. Bot.*, 1941, 28, 8713.
3. Sato, D., *Cytologia*, 1938, 9, 203.
4. Sharma, A. K. and Bal, A. K., *Ibid.*, 1956, 21, 329.
5. — and Ghosh, C., *Genetica Iberica*, 1954, 6, 91.
6. Venkatakrishnan, R., *Curr. Sci.*, 1975, 44, 19, 716.

PACHYTENE CHROMOSOMES IN AUTOTRIPLOID *BRASSICA CAMPESTRIS* VAR. *OLEIFERA* ($2n = 3x = 30$)

THE progeny of an autotetraploid *Brassica campestris* var. *oleifera* ($2n = 4x = 40$) contained an autotriploid with $2n = 3x = 30$. Since details of the chromosome couplement of the diploid and of the related autotetraploid species are available[12,13], an attempt is made to study the behaviour of associations of homologous chromosomes at pachytene with respect to particular chromosomes of the complement in this autotriploid. Quantitative details regarding the frequency and position of exchange of partners, the relative distribution of exchanges in the eu- and heterochromatic regions, the mean number and mean length of the pairing blocks were gathered with respect to the chromosomes 1, 4, 5, 7, 8 and 10. In general the details in the autotriploid were found to agree with those of the related autotetraploid plant presented elsewhere[13].

Although association is seen between all the three homologues at the heterochromatic segments flanking the centromere, in the euchromatic segments association is seen only between any two of them at any particular point the third one left unpaired as in the other triploids (Figs. 1 to 16). The mean number of exchanges ranged from 0 to 4. A positive correlation has been observed between the mean number of exchanges and the physical length of the chromosome (Fig. 17).

About 15·2% of the associations were with or without exchange of partners elsewhere on the chromosome but with association of heterochromatic segments of the three homologues at the centromeric region. The initial points of pairing as observed from the position of exchange of partners were found to fit to the 'Poisson' series, being distributed all along the length of the chromosome without any interference of one exchange on the chance formation of another. Although the occurrence of exchanges in the heterochromatic regions could not be seen due to lack of transparency in the material, this can however be inferred indirectly from the particular multivalent types met with at diakinesis.

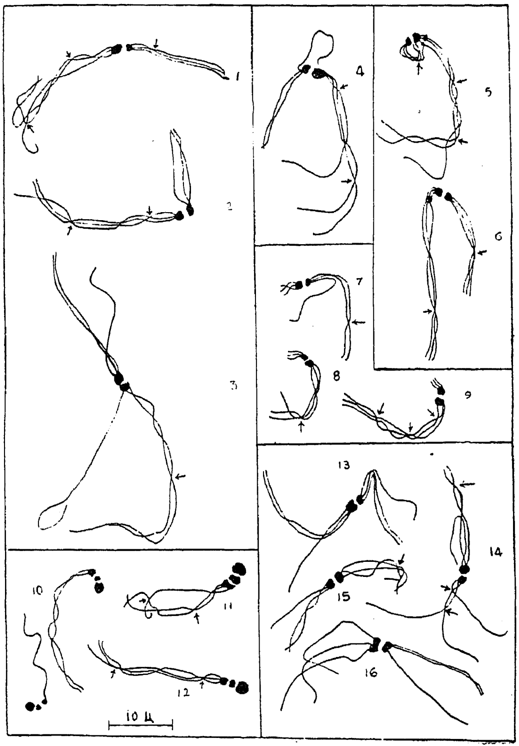
The trivalents noticed at diakinesis conformed to the 7th, 8th, 9th and 10th types of Darlington (1937) in the autotriploid plant (Table I). The 7th type

TABLE I

Frequency of different trivalent types at diakinesis in autotriploid Brassica campestris *var.* oleifera ($2n = 3x = 30$)

Trivalent type	7th	8th	9th	10th	Total nuclei
Number of nuclei	6	10	19	8	43

requires a minimum of one exchange in the centromeric region followed by 2 chiasmata in the two arms. The ninth type of trivalent was observed also in *B. campestris* ($2n = 20 + 2$) plant[4] tetrasomic for chromosome number 8. This chromosome is a short subterminal nucleolus organiser with a short arm of $0 \cdot 8\,\mu$ made of a proximal heterochromatic segment of $0 \cdot 3\,\mu$ and a distal hemispherical heterochromatic satellite of $0 \cdot 5\,\mu$ size. The ninth type requires a minimum of one chiasma in the two arms with one exchange in one of the arms. The 10th type of trivalent requires at least one exchange in each arm followed by an appropriate number of chiasmata at pachytene, proving thereby the occurrence of more than one exchange in the *Brassica* chromosomes. A quadrivalent of the 17th type of Darlington (1937) occurred in a greater frequency of the nuclei in autotriploid *B. campestris* var. *oleifera* ($2n = 3x = 30$), which requires a minimum of one exchange in the region including the centromere, with a minimum of 4 chiasmata in the two arms. It can be considered that although *Brassica* is having differentiated chromosomes, the exchanges of partners are not confined to the euchromatic regions alone and that they occur in the heterochromatic regions also as opposed to the generalisation made earlier by Venkateswarlu[11] based on the material available upto that time. Reddi[5,6] also reported an appreciable number of exchanges in the heterochromatic segments of tetraploid *Sorghum* species.



FIGS. 1 to 16. Associations of three homologous chromosomes belonging to chromosomes 1, 4, 5, 7, 8 and 10. Position of exchanges of partners indicated by arrow mark. Figs. 1 to 3. Chromosome 1. Fig. 4. chromosome 5. Figs. 5 and 6. Chromosome 7. Figs. 7 to 9. Chromosome 10. Figs. 10 to 12. Chromosome 8. Figs. 13 to 16. Chromosome 4.

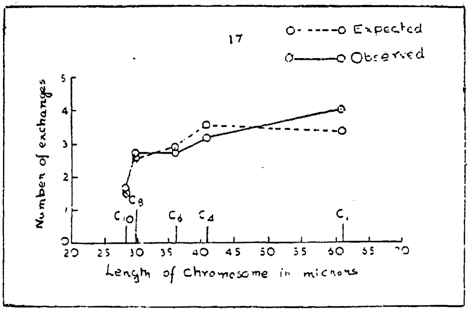


FIG. 17. Graph indicating the positive correlation between the frequency of exchanges and the physical length of chromosome in autotriploid *B. campestris* var. *oleifera* ($2n = 3x = 30$).

The mean number of the effective pairing blocks increased with increase in the mean length of the chromosome upto some length beyond which it decreased (Fig. 17) as has been noticed in other triploids and tetraploids[1–6,9,10,12]. The mean length of the chromosome which showed the maximum number of pairing blocks is $41\,\mu$ in both the autotriploid and autotetraploid *B. campestris* var. *oleifera*.

Thus the data in the autotriploid *Brassica campestris* var. *oleifera* agree in general with those seen in the autotetraploid plant presented elsewhere[13] showing that the pairing behaviour of pachytene chromosomes in the autotetraploids is an extension of that which takes place at the triploid level.

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---

1. Darlington, C. D., *Recent Advances in Cytology*, J. & A. Churchill, Ltd., 1937.
2. — and Mather, K., *Cytologia*, 1932, **4**, 1.
3. Gottschalk, W. Z., *Indukt. Abstam-u. Vererbungslehere*, 1955, **87**, 1.
4. Kamala, T., *Cytologia*, 1974, **39**, 253.
5. Reddi, V. R., *Genetica*, 1970 *a*, **41**, 321.
6. —, *Caryologia*, 1970 *b*, **23**, 295.
7. Röbbelen, G., *Chromosoma*, 1960, **11**, 205.
8. Stone, L. H. A. and Mather, K., *Cytologia* 1932, **4**, 16.
9. Tantravahi, R. V., The Bussey Inst., Harvard Univ., Cambridge, Mass., 1968, p. 1.
10. Venkateswarlu, J., *Ph.D. Thesis*, Cambridge Univ., Cambridge, England, 1950, p. 1.
11. —, Presidential address, Section of Botany 49th Indian Science Congress, 1962, p. 1.
12. — and Kamala, T., *J. Ind. Bot. Soc.*, 1971, **50 A**, 442.
13. — and —, *Ind. Jour. Genet. and Pt. Breed.*, 1974, p. 50.

SHORT SCIENTIFIC NOTES

Occurrence of *Ulocladium chartarum* on Mango in India

During March–April 1975, small (0·1 to 0·5 cm) irregular black lesions were observed on the inflorescence axis of mango varieties 'Maldah' and 'Sukul' at Muzaffarpur.

Fungus was isolated on Potato-dextrose agar slants at room temperature. The pure culture of the fungus was sent to C.M.I., Kew, England, and was identified as *Ulocladium chartarum* (Preuss) Simmons by Dr. M. B. Ellis (IMI, 200466). The pathogenicity of the fungus was established by inoculating the healthy axis of inflorescence under natural conditions. The symptoms produced were similar to those observed in nature. The fungus *Ulocladium chartarum* appears to be the first record on mango from India or abroad.

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September 15, 1976.

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A. K. SINHA.

Achlya hypogyna—A Notable Addition to Indian Aquatic Fungi

During the course of tax-ecological studies of aquatic fungi occurring in alkaline ('Usar') soils and water a species of *Achlya* has been isolated from an alkaline pond (pH 8·5) of village Devi Khera, situated in the vicinity of Lucknow-Rae-Bareli Road. The fungus was grown in sterilized distilled water on hempseed halves and identified as *A. hypogyna* Coker *et* Pemberton. However, this isolate differs from others described by various workers (Coker[1], Johnson[2], Ou[3]) in having thicker hyphae and non-papillate oogonia. This is the first report of the fungus from India.

Department of Botany,
Lucknow University,
Lucknow 226 007,
September 24, 1976.

J. N. RAI.
J. K. MISRA.

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1. Coker, W. C., *The Saprolegniaceae*, The Univ. of North Carolina Press, 1923, p. 201.
2. Johnson, T. W., Jr., *The Genus Achlya: Morphology and Taxonomy*, Univ. Michigan Press, Ann Arbor, 1956, p. 180.
3. Ou, S. H., *Sinensia*, 1940, 11, 427.

Trace Element Studies on Fungi. 1. Effect of Trace Elements on the Growth of Four Species of *Colletotrichum*

Effect of trace elements on the growth of large number of fungi have already been reported[1]. In the present investigation effect of 17 trace elements was studied on the growth of four species of *Colletotrichum*, namely, 1. *C. capsici* (Syd.) Butler and Bisby on *Capsicum annuum* L., 2. *C. hibisci* Poll. on *Hibiscus esculentus* L., 3. *C. indicum* Dastur on *Gossypium hirsutum* L. and *Colletotrichum* sp. Thind and Rawla on *Citrus limon* Burm., which were collected from Panjab and isolated by usual procedures. The procedure adopted for study was after Thind and Rawla[2]. The results of various experiments showed that Fe, Zn, and Mn are required for the growth of all these pathogens, Cu for the growth of *C. hibisci* and Mo for the growth of *C. capsici* and *C. hibisci*. The trace element requirements of these pathogens have not been reported so far.

Department of Botany,
Punjabi University,
Patiala (India), *October* 27, 1976.

S. S. SAINI.

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1. Sadasivan, T. S. and Subramanian, C. V., *Trans. Brit. Mycol. Soc.*, 1954, 37, 426.
2. Thind, K. S. and Rawla, G. S., *Proc. Ind. Acad. Sci.*, 1967, 66 B, 250.

A Note on the Occurrence of Genus *Microthamnion*

Microthamnion (order Chaetophorales, class Chlorophyceae) appears to be a rare genus which has not been recorded from India so far. While working on the microflora of Shillong, the present authors came across a form in the enrichment cultures of soil samples collected in October 1974 (supplemented with Bold's basal medium). The enrichment cultures with 5 g of soil sample in petridishes were incubated in a culture chamber illuminated with a fluorescent electric tube light (approximately 1000 lux intensity near the culture vessels at 25–30° C). This form was isolated in unialgal culture in Bold's medium under the cultural conditions mentioned above.

In culture, the thalli form a bushy appearance, possess almost a circular outline, grows as scattered patches of about 2 mm dia. on agar plates. Filaments lack a distinct main axis, are irregularly branched, cells are cylindric to clavate and almost uniformly broad, and the single plate-like chloroplast lacks a pyrenoid. On comparison with the known species the present alga resembles *M. kutzingianum* in all characters and measurements.

Department of Botany,
University of Allahabad,
Allahabad, *June* 4, 1976.

R. N. YADAVA.
D. C. PANDEY.

---

1. Deason, T. R. and Bold, H. C., University of Texas Publication No. 6022, 1960.
2. Printz, H., *Hydrobiologia*, 1964, **24**, 1–3, 284.

Sunflower Wilt—A New Disease from India

In 1973 *kharif* season, some of the fields of sunflower (*Helianthus annus* L.) grown at Parbhani were affected with wilt disease. The lower leaves changed from green to yellow and the plants toppled down and died. The fungus was also found in rootlets.

Fresh wilted plants were used for isolation on potato dextrose agar medium. The culture was purified by single hyphal tip method. Primary, secondary as well as tertiary septate hyphae grew independently with profuse branching. Microconidia were 0–2 septate and coniform or ellipsoidal in shape measuring $7 \cdot 25\,\mu \times 3 \cdot 15\,\mu$. Macro-conidia were sickle-shaped mostly 3–5 septate and were sparingly formed. The isolate was identified as *Fusarium oxysporum* Schlecht by Dr. Booth, Commonwealth Mycological Institute, Kew, Surrey, England. This fungus disease is recorded for the first time in India.

The pathogenicity was proved by soil inoculation method (variety Ec. 68415). The sunflower plants exhibited similar symptoms after inoculation as observed in naturally wilted plants. Repeated reisolations yielded *F. oxysporum* Schlecht.

College of Agriculture,
Parbhani (Maharashtra),
August 10, 1976.

B. N. GHODAJKAR.
N. M. GANACHARYA.
K. J. BINDU.
V. T. JADHAV.

Detached Leaf Culture, an Efficient Technique for Production of Viable Sclerotia of *Sclerotium rolfsii* Sacc.

Production of sclerotia by *Sclerotium rolfsii* was best on detached leaves of its host *Polyanthes tuberosa* floated on 10% sucrose solution kept in dark and incubated at 20–25° C. Large number of sclerotia of uniform size were observed at the periphery of the colony within seven days. Sclerotial formation in Carrot Potato Agar (CPA) and Potato Dextrose Agar (PDA) slants took more than sixteen days. Besides, the germination level of sclerotia produced on detached leaves was 70–80% in comparison to 20% viability of those from PDA and CDA slants. Pathogenicity tests with sclerotia produced in dlc showed typical symptoms of the disease on *P. tuberosa* within two weeks.

Division of Mycology
and Plant Pathology,
Indian Agricultural Research Institute,
New Delhi 110 012, *April* 21, 1976.

PRASANNA KUMARI PILLAI

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REVIEWS AND NOTICES OF BOOKS

Annual Review of Earth and Planetary Sciences (Vol. 4). Edited by Fred A. Donath, Francis G. Stehli and George W. Wetherill. (Annual Reviews, Inc., 4139 El Camino Way, Palo Alto, California 94306), 1976. Pp. 484. Price USA: $ 17.00; elsewhere: $ 17.50.

The present volume of the *Annual Review of Earth and Planetary Sciences* (Vol. 4, 1976) is a convincing evidence of the great diversity of these sciences. The review surveys different scientific investigations as the hydrogen loss rate from (planetary) atmospheres and the study of "Argillaceon Sediment Dewatering". What is most impressive is the vast scope for interdisciplinary research in these sciences. To illustrate, the article "On the generation of planetary magnetic fields" utilises a conceptual framework encountered quite frequently in the study of magnetic systems in stellar astrophysics, like pulsars. The technical difficulties associated with the understanding of the dynamics of charged fluid motions in electromagnetic fields are indeed similar. In fact, a surprising parallel with the technical problems associated even with non-linear field theories can be seen in the remarks on "Isotropic Turbulence".

Another article which highlights the interdisciplinary nature of the earth sciences is that on the hydrogen loss in planetary atmospheres. Apart from making very interesting reading, the article fully demonstrates the complexities as well as the range of concepts involved in such an investigation. Besides requiring expertise on such an interesting phenomenon as the solar wind, the subject requires a detailed understanding of the nature of proton collisions with hydrogen atoms bringing one to the realm of atomic physics. The presence of the earth's magnetic field adds its own distinctive charm by bringing in plasma physics too. The article deals with rather a novel (although relatively unknown to laymen), non-thermal mechanisms for hydrogen escape.

Apart from these two articles that I have specifically chosen to illustrate the interdisciplinarity of earth sciences, there are a number of others of interest. The article on SLAR imagery is an introduction to the observational methods in the study of plate tectonics. There is one on CH_4 and CO interaction which is a nice example of the subtleties involved in the study of atmospheres, and the artible is so thorough that it could well provide an interesting starting point for ecological studies.

N. D. Hari Dass.

Upper Air Sounding Studies. Vol. I: *Studies on Radiosonde Performance*. By A. H. Hooper; Vol. 11: *Manual Computation of Radiowinds.* By R. E. Vockeroth. (World Meteorological Organization Technical Note No. 140. WMO No. 394, Secretariat of the WMO, Geneva, Switzerland), 1975. Pp. 160 + 12, 18 figures.

As indicated, the Technical Note contains two reports prepared by two experts in the fields of radiosonde and radiowind observational systems, Volume I dealing with radiosonde compatibility, its assessment and with the results of radiosonde comparisons, while Volume II deals with the errors in manual computation of upper winds. The seeming disjointedness is due to the fact that these are two different reports prepared by the CIMO Working Group on Radiosonde and Radiowind Measurements established in 1965 and the Working Group on Radiosonde Instruments and Measurements established in 1969. These are all the same extremely valuable, as they show the several ways in which designers and operators of sondes can move towards their common goal of compatibility. The best that can be obtained from a radiosonde is that the mean values of data from a finite series of ascents and the data from individual ascents will have a statistical uncertainty which is small enough to avoid misleading the user. This is because even for sondes made to the same specifications, the errors vary from one occasion to the other and from one sonde to another and there is no means of determining the departure of a radiosonde from truth. The best that can be done is to see whether the errors of a radiosonde are the same from place to place. To assess this property of uniform of data, special sondes of significantly higher accuracy were designed by a number of national meteorological services. Even such reference sondes have an unknown residual error and it is only possible to assess the performance of the routine sonde within the limits of accuracy set by the reference sonde. Both laboratory and flight techniques were used to identify the individual sources of error and assess the overall performance of a routine sonde.

The various errors of a radiosonde are (*i*) the systematic error, (*ii*) the sonde error, and (*iii*) the random error. Systematic errors cannot be ascertained and are always present in radiosonde results. The "systematic difference" between the systematic error of one sonde design and the systematic error of another can, however, be determined. All the errors except the random errors vary in a regular way as the sonde ascends through the atmosphere.

The study shows that a statistical analysis of routine sounding data can best assess the performance of the sondes flown at the network of synoptic stations. Such analysis gives the overall quality of the data as received by the user, which is of most concern to him, as it provides continuing information upon current performance.

The report, outlines several different ways in which the compatibility of radiosondes can be studied. And if improved compatibility results by an application of the techniques reported, it will be a worthwhile achievement. It will also be apparent immediately as "data from radiosondes are, as it were, on display in a world-wide shopwindow, for all to see".

Part II of Volume I discusses the temperature reference sondes and their characteristics, the intercomparisons between them and the results that have been obtained. The work was carried out over many years and reflects the advances in the art that have been made since the concept of reference sondes was first evolved. However, the variability of the data is such that a repeat of the comparisons could well give substantially different results. Indeed, the magnitude of the variability is the most disappointing feature. Further work to reduce this variability would be necessary. It is also necessary to develop reference sondes for geopotential, pressure and humidity elements as well.

Part III of Volume I discusses the results of comparison of UK national and reference sondes. Apart from the evaluation of the alternative techniques for the comparison of radiosondes, the results bring out the difficulty in inferring the performance of radiosondes from such comparisons. The best method is the one of geopotentials provided by computer analysis. This reduces losses of data at high altitude during comparison trials and also enables comparisons of different sondes flown operationally at different places to be made. This is the latest tool available and will be useful in the future.

Volume II on Manual Computation of Radiowinds is an account of the extensive assessment of upper wind computation techniques made by CIMO Working Group on Radiowind Computation, established with the major task of developing principles and procedures for the computation of the required upper wind data that would have wide applicability to the major types of wind equipment.

The report discusses first the accuracy objectives and defines the measurement and computing errors. The general computing procedures are next described, of distance computation, computation of vector winds and radar and radiosonde height computation. Finally the last section describes the quality control of upper wind reports.

The selection of an appropriate practical system for computing upper level winds depends on the type and quality of the data available. The accuracy objectives have been laid down by the Working Group Accuracy Requirements in Aerological Soundings (WMO Technical Note 112). The author concludes that the accuracies achievable with manual radiowind computations are inadequate and the best of conventional manual computing procedures introduce errors greater than half the magnitude of measurement errors and often greater than the measurement errors. Inexpensive electronic calculators costing less than US $ 500 can be used to replace many slide rule and graphical calculations and can significantly reduce computing errors, while programmable desk calculators costing less than $ 5000 can reduce computing errors to negligible levels and greatly reduce the incidence of computing mistakes and at nominal costs provide the basis for a semi-automated computing system similar to those based on digital computers. Where cost can be justified. programmed digital computers should be interfaced with measuring equipment so that data transfer is automatic and measurement errors can be reduced by statistical filling procedures. All new systems being developed should have such computing capability included as an integral part of the systems in view of the increased accuracy and resolution that can be achieved. Further, detailed quality control procedures, using digital data processing, would be the best way to ensure that the computing system (like the measuring equipment) is operating close to its potential capabilities.

The reports provide a detailed and objective analysis of the errors involved on upper air measurements and suggest remedies and should be carefully studied by all scientists operating upper air networks in their countries.

6134/76. Published by S. R. S. Sastry, Manager. Current Science Association, Bangalore-560006
and Printed at The Bangalore Press. Bangalore-560018.

TRITIUM TRANSFER PATHWAYS IN THE AQUATIC PLANT *HYDRILLA VERTICELLATA*[*]

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Health Physics Division, Bhabha Atomic Research Centre, Trombay, Bombay 400 085

ABSTRACT

Rates of uptake and release of tritium by the aquatic plant *Hydrilla verticellata* were studied under controlled conditions. Significance of the persistence patterns of the aqueous and organic phases of tritium in the plant was established under different periods of exposure. The mean residence time for tissue-free-water-tritium was found to be 0·36 hours while the tissue-bound-tritium showed two components having mean residence time of 2·53 hours and 15·87 days respectively.

By reversing the pathways of tritium between the plant and the medium after long exposure times, the relative significance of the aqueous and organic phases of tritium in the plant and their persistence patterns have been established. It was also found that the water turnover rates in the plant was 0·028 ml/min/g.

INTRODUCTION

TRITIUM produced by the natural and artificial processes in the atmosphere is invariably present in the oxidized state as HTO and in this form it follows the hydrologic cycle, and entering into all hydrogen-containing compounds by isotopic exchange. Tritium transfer pathways in the ecosystems in general and in food chains in particular have attracted the attention of several workers[1-3]. Detailed investigations on the tritium persistence in the different compartments of the ecosystem have significant applications both in the field of radioecology and from environmental radiation point of view.

An attempt has been made in the present work to generate data on the parameters governing the fixation of tritium in the organic phase. The mean residence times have been calculated on the basis of whole tritium values obtained from samples exposed for different periods of times under different conditions of exposure. By this method the significant fraction of tritium in the plant is gradually changed from tissue-free-water-tritium (TFWT) to tissue-bound-tritium (TBT) labile component and later to TBT non-labile component.

MATERIALS AND METHOD

About 2 kg of *Hydrilla verticellata* obtained from the Taraporewalla Aquarium, Bombay, were exposed to varying periods of time in a laboratory aquarium of approximately 25 l capacity, under controlled humidity and temperature conditions. The medium water was changed at a fixed time schedule depending on the type of investigations. The plants were constantly exposed to 40 W fluorescent lighting throughout the period of study.

The exposure time of the aquatic plants in the active media was varied in each of the experiment in order to determine the period of time required for effective tritium transfer between the two main hydrogen pools of the system, *viz.*, the plant water and the organic fraction in the plant.

The periodicity of sampling varied from 15 mts to 24 hrs depending upon the status of exposure of the plant-system. The wet sample collected at random was dried using a filter paper taking care not to break their stems while removing the adhering water droplets. The wet and lyophilised tissues were burnt separately in an oxygen atmosphere in order to extract the activity contained in HTO or HT[4]. The activity in the extracts is then counted in a Liquid Scintillation Spectrometer (Packard Model 2211) applying appropriate corrections for quenching.

Hydrogen content of the lyophillised tissue was estimated by burning the sample and absorbing the steam in $MgClO_4$, using conventional chemical apparatus.

Exposure schedule.—F. L. Harrison *et al.*[3], describe the procedures required to fix preferentially the TBT in certain marine animals on the basis of a mathematical model which takes into effect all the possible tritium pathways in a total aquatic environment. The same model was used here to a major extent with only minor modifications for application to aquatic plants. The aquatic plants are exposed to active media for a certain period and are transferred to inactive media in stages thereby eluting the different components of tritium compounds in the plant. Periodic sampling was carried out on the whole plant for total tritium content in order to identify the nature of release patterns. The investigations are divided into a number of batches of experiments.

A preliminary run indicated the fast water turnover rates of these plants. The tritium concentration

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[*] Work carried out under the International Atomic Energy Agency Research Contract No. 1297/R2/RB.

values of the pool water was increased from 10^4 dpm/ml to 10^6 and 10^8 dpm/ml in order to have sufficient concentration in the TBT phase.

The plants were exposed to 8 hours in an active medium to determine the TFWT uptake pattern and then were transferred to inactive medium to determine the TFWT and labile TBT release patterns. These studies were continued upto 80 hours.

The next batch of experiments on the plant covered three different phases of investigations :

Phase 1.—The plants were immersed in a tritiated water medium (concentration of about 10^8 dpm/ml) for a period of 18 days during which period the uptake pattern and equilibrium behaviour under controlled conditions were studied. The medium water was also regularly monitored, in order to correlate the saturation activity levels of the plant and the medium, as well as to study the loss of tritium from the aquarium due to evaporation.

Phase 2.—The plants exposed to tritium concentrations as per the schedule in phase 1 were then transferred to an inactive pool and the investigations continued for 42 days. During this phase the TFWT release pattern as well as labile and non-labile tritium release patterns were studied. The pool water concentrations were found to have increased during this period reaching an equilibrium value. This is attributed as due to tritium releases from the aquatic plant to the medium water.

Phase 3.—The plants were transferred at this juncture to a freshwater aquarium for a second time. The **TBT** release patterns were found to be the most prominent one during this period.

RESULTS AND DISCUSSION

Figure 1 presents the results of the rate of uptake and release of tritium for short intervals of time. The aquatic plants reached an equilibrium concentration corresponding to 80% of the medium water concentration, within a period of one hour, when normalised for moisture content of the plant (0·76 w/w). A least square analysis of the results showed three components of release pattern. The mean residence times corresponding to TFWT phase and TBT labile phase were found to be 0·36 h and 2·53 h respectively.

The water turnover rate of the plant was determined on the basis of the steady-state tracer kinetics[5]. Under equilibrium conditions

$$S_m \cdot R = \lambda \cdot S_s$$

where S_m and S_s are specific activity of tritium in the medium (cpm/ml) and the sample (cpm/wet.g), R is the rate of uptake (ml/min.) and λ is the inverse of the tritium mean residence time in minutes.

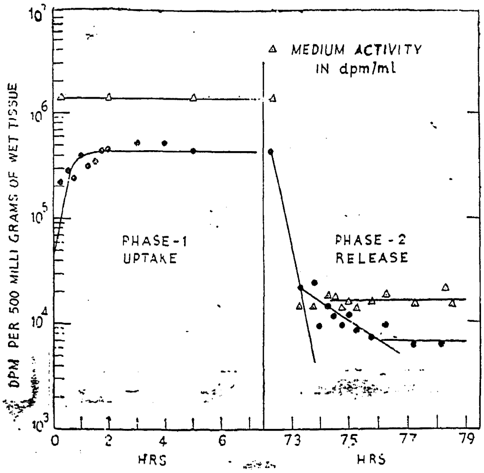

FIG. 1. Water turnover rate and TFWT release pattern

Since the mean residence time values for TBT are large compared to those of TFWT, R is calculated as 0·028 ml/min./g corresponding to a tritium mean residence value of 0·36 h.

Figure 2 shows the three phases of tritium transfer pathways observed in the experiments. Phase 1 has an uptake pattern identical to that in Fig. 1, except for the evaporation losses shown in the medium concentration values (as well as those shown by the corresponding aquatic plant samples). In phase 2, the two components of tritium releases are observed. The first one shows an extremely rapid rate of release corresponding to that of TFWT and TBT labile components. The second component shows the long-lived TBT non-labile tritium in the aquatic plants. The mean residence time for this component is found to be 15·87 days indicating that the organic phase transfer mechanism has an extremely slow rate constant compared to the other phases. Under equilibrium conditions, when the tritium concentrations of the plant tissue were normalised, the medium water showed values of specific activity lower than those for the plant tissue for a few days confirming the fact that pathways (of tritium) have been reversed due to the preferential incorporation of tritium in the organic phase of the plant when immersed in an inactive medium.

In phase 3, the above phenomenon is highly pronounced due to the fact that the only major component of tritium left behind is the non-labile one. The mean residence time was once again found to be same, *viz.*, 15·87 days.

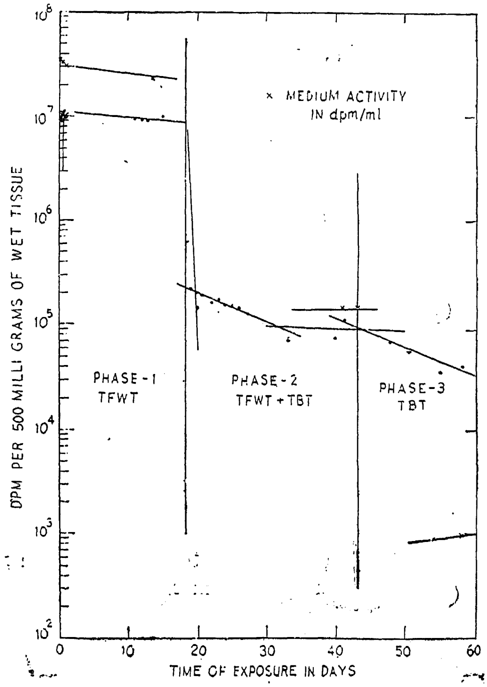

FIG. 2. Tritium uptake and release pattern or long periods of exposure.

Figure 3 shows the schematics of the tritium transfer pathways in the aquatic plant as reported in the experimental observations (*vide* Figs. 1 and 2) during different phases of the experiments.

The relative fractions of tritium incorporated in the three phases of the bound and free forms of tritium in the aquatic plant were found to be 100 : 3·27 : 0·77. This shows that 3·14% of tritium is in the labile TBT form and 0·74% is in the non-labile TBT form. This could be the reason for the necessity to have high concentrations of medium water in order to obtain a preferential fixing of TBT in the aquatic plant. The values obtained for the different fractions of tritium (bound and free) in the aquatic plant *Hydrilla verticellata* in the above study agree well with the values obtained by H. D. Bruner[6]. He has obtained similar orders of magnitude in his study of the specific activity values for different forms of bound and free tritium in phytoplankton, zooplankton and certain aquatic plants.

The hydrogen content of the lyophillised tissue of the aquatic plant was found to be 7·26%. The moisture content of the plant being 0·76 w/w, it can be seen that the available hydrogen in organic phase is 1·74%. However the total TBT fraction as determined by uptake measurements was found to be 3·85%. This variation in values can be attributed to either or both of the following reasons.

(*i*) The value of hydrogen content in organic phase can be on the lower side, due to possible losses of certain volatile substances during lyophillisation.

(*ii*) The TBT fraction as determined by the analysis of total ^{3}H content estimations of the aquatic plant can show a slightly higher value due to possible interferences from TFWT fractions. In order to circumvent this the mathematical treatment has to be enlarged so as to determine the extent of such interferences.

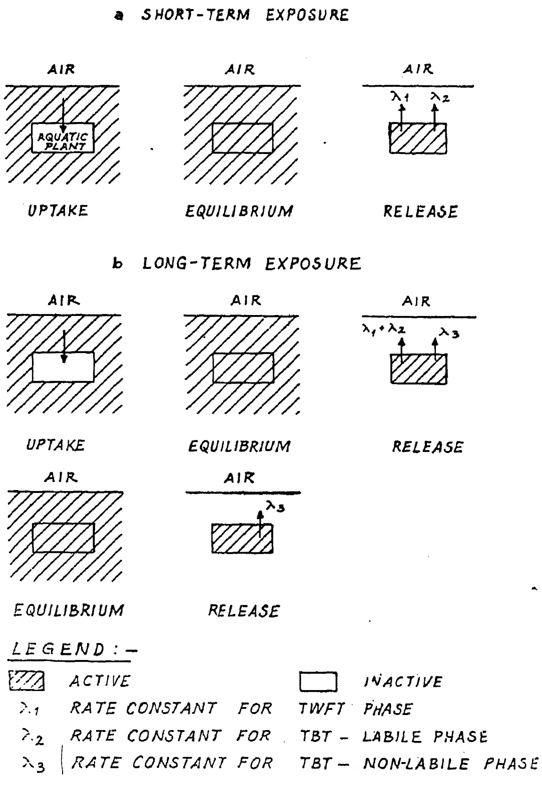

FIG. 3. Schematics of the tritium transfer pathways.

CONCLUSION

The aquatic plant, *Hydrilla verticellata,* shows three distinct mean residence time values for tritium, one relating to aqueous phase and two corresponding to the organic phase. It is possible to determine these components by estimating the total tritium content of the plant samples at different periods of time after a certain type of exposure schedule. Further, it has been observed that the tritium pathway can be reversed between the medium and the plant, after long exposure times.

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1. Anspaugh, L. R. *et al.*, *Tritium*, Ed. A. Alan Moghissi and Melvin W. Carter (Messenger Graphics, Phoenix, Arizona and Las Vegas, Nevada), 1973, pp. 405.
2. Kirchmann, R. *et al.*, *Ibid.*, 1973, p. 341.
3. Harrison, F. L. *et al.*, *Ibid.*, 1973, p. 363.
4. Vaze, P. K., *et al.*, *A Simple Technique for Determination of Different Fractions and Total Tritium Content of Sample for Field Use*, IAEA Coordination Meeting, Mol. Belgium (April, 1975).
5. Soman, S. D., and Krishnamoorthy, T. M., *Curr. Sci.*, 1973, 42 (13), 453.
6. Bruner, H. D., *Tritium*. Ed. A. Alan Moghissi and Melvin W. Carter (Messenger Graphics, Phoenix, Arizona and Las Vegas, Nevada), 1973, p. 303.

ULTRASONIC RELAXATION IN ETHYL SALICYLATE

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ABSTRACT

Measurements of ultrasonic absorption have been made by pulse technique in liquid ethyl salicylate and also in its solutions with *m*-xylene and nitrobenzene as solvents. Results are analysed on the basis of single relaxation mechanism. Relaxation frequency is found to be independent of the concentration of ethyl salicylate in both the solvents. Results are explained on the basis of rotational isomerism in ethyl salicylate.

SEVERAL investigators[1-3] have reported ultrasonic relaxation effects in esters of monocarboxylic acids. Relaxation in OH substituted esters of carboxylic acids has not been thoroughly investigated. These esters from a separate class in that they exhibit intramolecular hydrogen bonding. Murthy and Rao[4] have reported the relaxation in liquid methyl salicylate which was attributed to rotational isomerism. In this note measurements of ultrasonic abosorption in pure liquid ethyl salicylate and also in solutions of ethyl salicylate with *m*-xylene and nitrobenzene are presented.

Measurements of absorption and velocity have been made by pulse technique in the frequency range 4 to 15 MHz. Liquids used are of high grade purity and have been distilled before use.

Results of absorption have been analysed on the basis of a single relaxation mechanism

$$\frac{\alpha}{f^2} = B + \frac{A}{1 + \left(\frac{f}{f_r}\right)^2}$$

where A and B are constants of relaxation and f is the characteristic frequency of relaxation.

The excess absorption per wavelength ($\alpha'\lambda$) at any frequency is calculated from

$$(\alpha'\lambda) = \left(\frac{\alpha}{f^2} - B\right) f C$$

Maximum excess absorption per wavelength μ_m is computed from

$$\mu_m = \frac{1}{2} A f_r C$$

where C is the velocity of sound.

TABLE I

Ultrasonic absorption and relaxation parameters in ethyl salicylate

Temp. °C	v m/sec	f Mc/sec	$10^{17} \times \alpha/f^2$ cm⁻¹ sec	$10^2 \times \alpha'\lambda$ *db*	10^{17} A cm⁻¹ sec²	f_r Mc/sec	10^{17} B cm⁻¹ sec²
30	1360	4·44	1300	6·7	2500	3·9	30
		7·20	570	4·6			
		10·10	360	3·9			
		13·00	225	3·0			
40	1300	4·44	1365	6·5	2000	4·9	60
		7·20	680	5·0			
		10·10	430	4·2			
		13·00	320	3·8			
50	1240	4·44	1400	6·4	1666	6·3	65
		7·20	800	5·7			
		10·10	490	4·6			
		13·00	410	4·8			

The computed values of α/f^2, $(\alpha'\lambda)$, f_r and μ_m are given in Table I for ethyl salicylate and in Tables II and III for solutions in *m*-xylene and nitrobenzene respectively.

TABLE II

Ultrasonic absorption and relaxation parameters of ethyl salicylate in m-*xylone*

Volume %	v m/sec	f Mc/sec	$10^{17} \times \alpha/f^2$ cm^{-1} sec^2	$10^2 \times \alpha'\lambda$ db	10^{17} A cm^{-1} sec^2	f_r Mc/sec	$10^3 \times \mu$m	10^{17} B cm^{-1} sec^2
17	1308	4·44	270	1·03	400	4·04	1·01	65
		7·20	160	1·78				
		10·10	120	0·63				
		13·00	105	0·59				
33	1319	4·44	480	2·14	6·6	4·10	1·64	60
		7·20	235	1·44				
		10·10	160	1·46				
		13·00	120	0·89				
50	1331	4·44	680	3·21	1110	4·00	2·90	55
		7·20	285	1·91				
		10·10	215	1·87				
		13·00	150	1·43				
65	1345	4·44	880	4·33	1508	3·92	3·98	45
		7·20	400	2·99				
		10·10	260	2·54				
		13·00	170	1·90				
100	1360	4·44	1300	6·66	2500	3·87	6·60	30
		7·20	570	4·59				
		10·10	360	3·94				
		13·00	225	2·00				

TABLE III

Ultrasonic absorption and relaxation parameters of ethyl selicylate in nitrobenzene

Volume %	v m/sec	f Mc.sec	$10^{17} \times \alpha/f^2$ cm^{-1} sec^2	$10^2 \times \alpha'\lambda$ db	10^{17} A cm^{-1} sec^2	f_r Mc/sec	$10^3 \times \mu$m	10^{17} B cm^{-1} sec^2
17	1437	4·44	320	1·21	287	4·02	0·83	100
		7·20	190	0·81				
		10·10	140	0·50				
		13·00	125	0·40				
33	1429	4·44	500	2·33	800	4·10	2·27	75
		7·20	260	1·64				
		10·10	185	1·37				
		13·00	140	1·04				
50	1421	4·44	700	3·44	1250	3·85	3·38	65
		7·20	335	2·43				
		10·10	230	2·04				
		13·00	165	1·59				
66	1413	4·44	890	4·50	1666	3·70	4·28	50
		7·20	410	3·13				
		10·10	270	2·67				
		13·00	185	2·12				
100	1360	4·44	1300	6·66	2500	3·87	6·60	30
		7·20	570	4·59				
		10·10	360	3·94				
		13·00	225	3·00				

Variation of log (f_r/T) with 1/T is shown in Fig. 1 for pure ethyl salicylate. The observed linear variation with negative slope is consistent with the theory proposed by Davies and Lamb[5] for rotational isomerism. Estimated value of activation energy of the rate determining mechanism is 5 K.cals/mole.

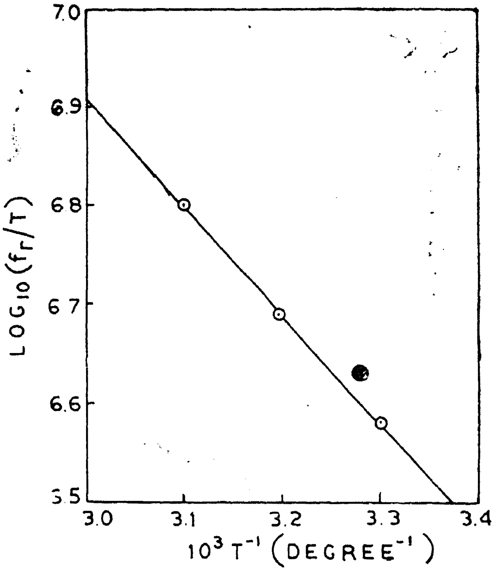

FIG. 1. Plot of Log_{10} (f_r/T) *vs*. T^{-1} for ethyl salicylate.

From Tables II and III it is apparant that the relaxation frequency f_r of ethyl salicylate is not significantly altered by the foreign environment of either *m*-xylene or nitrobenzene. Polar and non-polar solvents have no influence on the relaxation, except for providing a dilution effect on relaxation strength. It can therefore be concluded that the mechanism of relaxation in ethyl salicylate is of an intramolecular origin.

ACKNOWLEDGEMENT

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1. Hall, D. N. and Lamb, J., *Trans. Faraday Society*, 1959, 55, 784.
2. Subrahmanyam, S. V. and Piercy, J. E., *J. Acoust. Soc. Am.* 1965, 37, 340.
3. —, and —, *J. Chem. Phys.*, 1965, 42, 1475.
4. Sree Ramamurthy, J. and Ramachandra Rao, B., *J. Acoust. Soc. India,* 1973, 1, 170.
5. Davies, R. O. and Lamb, J., *Proc. Phys. Soc.* London, 1959. 73 B, 767.

SYNTHESIS OF 3″-METHYL-FURO-(5″, 4″: 7, 8)-FLAVONES

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FUROFLAVONE, karanjin and furocoumarins, psoralene, angelecin and xanthotoxin are reported to possess fish-toxic and photodynamic properties[1-3]. Several methyl substituted furocoumarins and furoflavones are also reported to possess similar physiological properties[4-6]. 3′, 4, 8-Trimethylpsoralene, available in large quantities by synthesis, is the most effective photodynamic agent[7]. 3″-Methyl-furo-(5″, 4″: 7, 8)-flavones, considered to be analogous of karanjin, do not seem to have been synthesized earlier. In this communication we report their synthesis, spectral characteristics and fish-toxicity.

7-Hydroxy-(I *a*)[8], 7-hydroxy-3-methyl-(I *b*)[9], 7-hydroxy-3-methyl-3′, 4′-dimethoxyl(I*c*)[9] and 7-hydroxy-3-phenyl(I *d*)[10]-flavone were condensed with chloroacetone by heating in acetone-potassium carbonate medium to give the corresponding 7-acetonoloxyflavones (II *a–d*). Their melting points, ir and uv data are given in Table I and the nmr data are given in Table II. The cyclodehydration of acetonoloxybenzenes with polyphosphoric acid, leading to the formation of 3-methylbenzofurans, was reported earlier[11]. The 7-acetonoloxyflavones (II *a–d*) were heated in polyphosphoric acid for 2 hours at 110–120°. In each case a pale yellow semi-solid was obtained. The semi-solids were subjected to chromatography, resulting in the isolation of (1) cyclized product ~30%, (2) unconverted starting compound ~50%, and (3) the corresponding 7-hydroxyflavones ~20% in each case. The melting points, ir and uv data of the cyclized products III *a–d* are given in Table I and their nmr data are given in Table II. The data suggest that the cyclized products possess 3-methyl-furo-(5″, 4″: 7, 8)-flavone structure (III *a–d*). In the ir spectra of furoflavones the frequency appearing around 1630 cm^{-1} is due to the flavone carbonyl group. The carbonyl frequency found around 1725 cm^{-1} in 7-acetonoloxyflavones (II *a–d*) due to acetonoloxy carbonyl disappears in the furoflavones. In their nmr spectra, a doublet appears in the region 2·45–2·65 δ (J = 1 Hz) assignable to the C3″-methyl group and the splitting is due to the adjacent C2″ proton. An AB doublet appears in the region 8·20–8·24 δ (J = 9–10 Hz), characteristic

TABLE I

*Mp, ir, uv and fish-toxicity data of 7-acetonoloxyflavones and the corresponding 3″-methyl-furo-(5″, 4″: 7, 8)-flavones**

Compound	mp °C	ir ($CHCl_3$) cm^{-1} Acetonol-oxy carbonyl	ir ($CHCl_3$) cm^{-1} Flavone carbonyl	uv MeOH max. nm (Log ϵ)	Fish-toxicity at 20 ppm turning time Mts. Sec.
II *a***	179	1730	1630	222 (4·04), 252 (4·04) and 306 (4·17)	10–00
II *b***	132	1725	1625	236 (4·25) and 300 (4·26)	4–30
II *c***	95	1730	1620	224 (4·31) and 308 (4·31)	12–00
II *d***	170	1725	1630	232 (4·38), 258 (4·23) and 306 (4·25)	35–00
III *a***	130	..	1635	224 (4·30), 256 (4·32), 264 (4·33) and 300 (4·10)	Not toxic within 45 miutes
III *b*†	190	..	1630	222 (4·28), 254 (4·38) and 310 (3·99)	do.
III *c*†	184	..	1630	222 (4·36), 244 (4·45), 248 (4·45) and 316 (4·23)	do.
III *d*†	215	..	1630	224 (4·56), 250 (4·57), 258 (4·57) and 308 (4·15)	do.

* Satisfactory elemental analyses were obtained for all the compounds.
** Solvent of crystallization: Benzene-Petroleum ether (60–80°).
† Solvent of crystallization: Benzene.

TABLE II

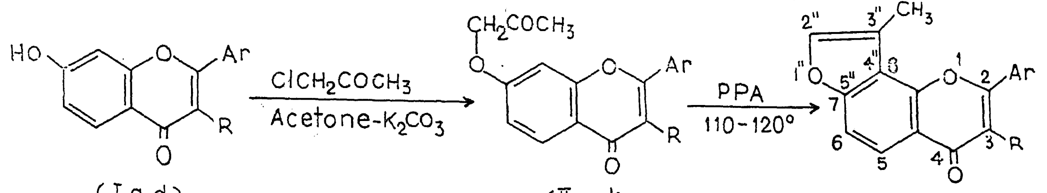
Nmr spectral data of 7-acetonoloxy-, and 3″-methyl-furo-(5″, 4″: 7, 8)-flavones

Solvent ($CDCl_3$) 60 MHz, δ values, (J in Hz)

Compound	C3″ (d)	$-O-CH_2-$ (s)	$-CO-CH_3$ (s)	C3 substituent (H or CH_3 or Ph)	C5	C2″, C2 phenyl, C6 and C8 (m)	Methoxyls (s)
II *a*	..	4·70	2·18	6·85 s (H)	8·25 (J = 9)	7·00–7·50 (7H)	..
II *b*	..	4·70	2·10	2·30 s (CH_3)	8·25 (J = 10)	6·75–7·70 (7H)	..
II *c*	..	4·62	2·12	2·28 s (CH_3)	8·15 (J = 9)	6·70–7·40 (5H)	3·9 (6H)
II *d*	..	4·72	2·30	6·90–7·40* m (C_6H_5)	8·30 (J = 10)	6·90–7·40* (12H)	..
III *a*	2·65 (J = 1)	..	..	6·90 s (H)	8·20 (J = 9)	7·30–7·75 (7H)	..
III *b*	2·45 (J = 1)	..	..	2·24 s (CH_3)	8·24 (J = 9·5)	7·30–7·90 (7H)	..
III *c*	2·55 (J = 1)	..	..	2·35 s (CH_3)	8·23 (J = 9)	7·00–7·63 (5H)	4·05 (6H)
III *d*	2·52 (J = 1)	..	..	7·18–7·60* m (C_6H_5)	8·20 (J = 10)	7·18–7·60* (12H)	..

* C3 Phenyl protons merged with other aromatic protons.

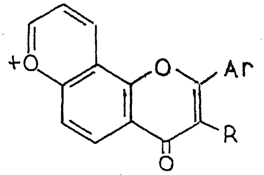
of C5 proton of flavones[12], indicating that there is a proton at C6 position. The C2″ and C6 protons merge with the other aromatic protons of side phenyl nuclei. From this it can be inferred that the mode of cyclization is 7 : 8 positions of flavone skeleton rather than 6 : 7 positions. This is in agreement with the earlier observation that electrophilic substitution reactions in 7-hydroxyflavones occur exclusively at position-8[13]. In the mass spectra, the molecular ion peak is quite abundant in all the furoflavones III *a–d*. The base peak is due to M–1 ion (IV *a–d*) formed as a result of the facile loss of a hydrogen from the C3″-methyl group followed by a rearrangement to give a stable ring expanded chromenyl ion[14]. A less intense M–CO ion (V *a–d*) and intense ions due to retro-Diels-Alder fission (VI *a–d* and VII *a–d*) are also noticed[15].



(I a-d) (II a-d) (III a-d)

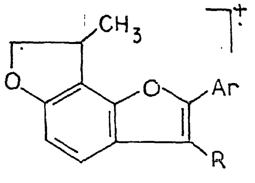
(a) R = H, Ar = Ph
(b) R = CH$_3$, Ar = Ph
(c) R = CH$_3$, Ar = $C_6H_3(OCH_3)_2$
(d) R, Ar = Ph

M$^{+\cdot}$(IIIa) 276 (60%)
(IIIb) 290 (64%)
(IIIc) 352 (72%)
(IIId) 350 (34%)



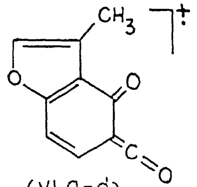
(IV a-d)

(a) m/e 275 (100%)
(b) " 289 (100%)
(c) " 351 (100%)
(d) " 349 (57%)



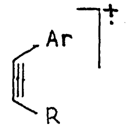
(V a-d)

(a) m/e 248 (5%)
(b) " 262 (4%)
(c) " 324 (4%)
(d) " 322 (11%)



(VI a-d)

(a) m/e 174 (27%)
(b) " (24%)
(c) " (28%)
(d) " (100%)



(VII a-d)

(a) m/e 102 (15%)
(b) " 116 (17%)
(c) " 178 (47%)
(d) " 176 (100

7-Acetonoloxyflavones II *a–d* at 20 ppm are fairly toxic to fish species *Barbus ticto* (Table I) when tested adopting the procedure of Krishnaswamy and Seshadri[2]. At the same concentration the furoflavones III *a–d* did not show any toxicity within 45 minutes. Screening of these compounds for photodynamic activity is in progress.

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1. Seshadri, T. R. and Sood, M. S., *Curr. Sci.*, 1963, **32**, 195.
2. Krishnaswamy, B. and Seshadri, T. R., *Proc. Indian Acad. Sci.*, 1942, **16 A**, 231.
3. Späth, E. and Kuffner, F., *Monatsh.*, 1936, **69**, 75.
4. Fowlks, W. L., Griffith, D. G. and Oginsky, E. L., *Nature*, 1958, **181**, 571.
5. Pathak, M. A., Fellman, J. B. and Kaufman, K. D., *J. Invest. Dermatol.*, 1960, **35**, 165.
6. Chibber, S. S., Ganguli, A. K., Mukerjee, S. K. and Seshadri, T. R., *Proc. Indian Acad. Sci.*, 1957, **46 A**, 19.
7. Kaufman, K. D. and Worden, L. R., *J. Org. Chem.*, 1961, **26**, 4721.
8. Jain, A. C., Mathur, S. K. and Seshadri, T. R., *J. Sci. Industr. Res.*, 1962, **21 B**, 214.
9. Srimannarayana, G. and Subba Rao, N. V., *Indian J. Chem.*, 1968, **6**, 696.
10. Gupta, V. N. and Seshadri, T. R., *J. Sci. Industr. Res.*, 1957, **16 B**, 116.
11. Weinheimer, A. J. and Washecheck, P. H., *Tetrahedron Lett.*, 1969, p. 3315.
12. Mabry, T., Markham, K. and Thomas, M., *The Systematic Identification of Flavonoids*, Springer-Verlag, Berlin, 1970, p. 274.
13. Dean, F. M., *The Total Synthesis of Natural Products*, Vol. I (Ed. J. ApSimon) Wiley-Interscience, New York, 1973, p. 508.
14. Willhalm, B., Thomas, A. F. and Gautschi, F., *Tetrahedron*, 1964, **20**, 1185.
15. Porter, Q. N. and Baldas, J., *Mass Spectrometry of Heterocyclic Compounds*, Wiley-Interscience, New York, 1971, p. 171.

FIRST ALL-INDIA SEMINAR ON ENVIRONMENTAL GEOSCIENCES

The Department of Geology, Bangalore University and the Indian Institute of Geological Education jointly organised the First All-India Seminar on Environmental Geosciences. The objectives of the seminar were: (1) to initiate firm base for the subject of Environmental Geosciences which are new to this country and so vital for the mankind, (2) to group the present available knowledge on the subject with the help of the scientists engaged in the field, (3) to formulate a sound programme of Environmental Geology by constructing a curriculum for a course through group discussion and (4) to suggest specific ideas for planners and policy makers. The deliberations were concerned with different issues of environmental problems of vital importance.

The seminar was held in Bangalore from 8th to 10th August 1976 and was attended by 100 delegates from all over the country. The deliberations were conducted in five sessions and one workshop. Thirty-two papers were received dealing with the implications of Environmental Geosciences on different aspects.

In the first session Dr. T. Thimaiah stressed the importance of scientific approach on dumping of mining waste and its impact on environment through a key note address. R. Srinivasan and B. L. Srinivas highlighted the concept of land reclamations of derelict mined lands. The paper by R. H. Sawkar and V. N. Vasudev dealt with the disposal of copper mine waste and pollution control.

In his key note address to the second session Dr. K. V. Raghava Rao mentioned the impact of natural environmental forces upon man. The toxic chemicals in mineral environment and the continued application of fertilizers leading to the infertility of soil was pointed by J. S. Venugopal. The paper by J. C. V. Sastry and others dealt with the industrial pollutants through a case study of Mandya National Paper Mills.

Prof. S. Sambe Gowda, while delivering his key note address to the third session, brought out the importance of environment on the evolution of life in the past and the usefulness of this knowledge in predicting the future environment. Dr. V. V. Pathy and others dealt with the compositional control of biological factor of the sedimentary environment in Kaladgi basin of Northern Karnataka. Graphical analysis of EH and PH studies of Gurmutkal area and its correlation with the ecosystem of faunal and floral remains was presented by T. V. Shivarudrappa.

Sri J. P. Srivastava laid stress on the need for co-operative efforts of the engineers, town planners and architects while delivering the key note address to the IV session. An attempt to decipher environmental geology factors of the twin cities of Ahmedabad–Gandhinagar was presented by R. S. Shenoi. The importance of geology in urban and industrial planning of towns with special reference to Jaipur and Alwar was presented by P. N. Mehta and R. B. Nag. K. C. C. Raju dealt with the urban and rural water development schemes through a case study of Anantapur District. Fluoride and fluorosis from ground water sources and its effect on health was presented by K. N. Chandrashekara.

The last session of the seminar was earmarked for a workshop to frame a curriculum for a course in "Environmental Geosciences" through group discussion. A book exhibition was also arranged in this connection.

The seminar was inaugurated by Sri. K. Rajagopalaswamy, President, Mineralogical Society of India. Prof. M. Shadakshara Swamy, Principal, Central College, Bangalore, in his presidential address spoke on the importance of environmental geosciences. He stressed the need for co-operative action by chemists, engineers and environmental geologists in evolving a course. The authors of this report hope that this 'First All-India Seminar on Environmental Geosciences' will lay a firm foundation for the development of this new branch Earth Science, a branch which has Geology as its foundation.

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LETTERS TO THE EDITOR

A NOTE ON THE EQUATORIAL STRATOPAUSE

THE temporal variations of stratospheric parameters studied[1–3] revealed large variations in temperature and windspeeds at the stratopause. Schwentek[4] showed a good solar influence on the stratospheric temperatures of West Berlin for the winter atmosphere of the sunspot cycle, 1957–70. The recent studies of Ramakrishna and Seshamani[5] point out a strong correlation between mesospheric temperatures at Thumba and 10·7 cm solar flux 1970–71.

The diurnal variations in temperature of the stratopause is controversial. Beyers and Miers[1] observed stratopause temperatures to be at minimum at local midnight and maximum at local noon for whitesands (32° North). However, Theon *et al.*[3], reported for the stratospheric region 30 to 60 km a large increase of temperature during 2000–2400 hrs and abrupt cooling during 01:00–05:00 hrs Wallops island (38° North).

In view of these various controversies a study is attempted in the present paper to understand equatorial stratopause during day and night and to study the solar influence on its parameters. The stratopause is a level of maximum in the vertical temperature structure.

The data obtained from the M-100 type Meteorological rocket firings made regularly at 20:00 hrs at Thumba, beginning in January 1971, give the altitude profile of the wind, temperature, pressure and neutral density upto an altitude of about 90 km. The earlier flights of January 1971 give some data during daytime. These data are used for the day and nighttime studies and those of January 1972–December 1973 for correlative studies. The present study is mainly confined to temperature and density variations.

The average of the two daytime flight measurements of temperature at 10:00 hrs (20 January 1971) and 10:22 hrs (30 January 1971) is taken as the daytime representative profile and this is shown in Fig. 1. Another profile averaged from the three flight measurements made at 23:00 hrs (6 and 20 January 1971) and 23:05 hrs (13 January 1971) is also shown to represent the nighttime profile. The temperature profiles show a large increase, through a region of about 15 km from day to night conditions around the stratopause. The maximum temperature difference of 12·3° C was observed at 40 km altitude.

The large diurnal variations in temperature led to the examining of the solar influence on the stratopause. Hence, correlations between stratopause and solar parameters are studied, and the temperature and density data of the nighttime flights around 20:00 hrs over a two-year period are analysed. This period (January 1972–December 1973), during which 90 flights were made, also coincides with one biennial cycle[4] which is exhibited by most of the stratospheric parameters. The solar parameters of the corresponding day are taken to study the correlation. The correlative data are taken from *Solar Geophysical Data Bulletin* published by the NOAA. Choosing the solar data of the corresponding day is justified as it was already shown that the solar influence is felt in less than a day's period at mesospheric heights[5]. The solar influence will be felt with a lesser delay at decreasing altitudes.

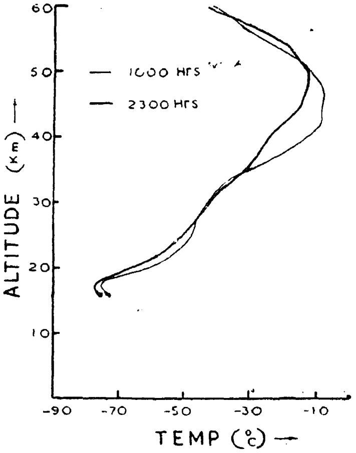

FIG. 1. Altitude profiles of temperature for Thumba averaged over two daytime and three nighttime flights of January, 1971.

The cross-correlations coefficients calculated with the Zurich sunspot number and 10·7 cm solar flux of Ottawa are given in Table I with their corresponding

TABLE I

Correlation coefficients evaluated between stratopause and solar parameters

Correlations between	No. of observations	Correlation coefficient	Significance level*
Temperature *vs.* Sunspot number	90	0·37	<0·01
Temperature *vs.* Solar radio flux	90	0·28	<0·01
Neutral density *vs.* Sunspot number	90	0·09	..
Neutral density *vs.* Solar radio flux	90	0·07	..

* < 0·01 indicates high correlation, .. poor correlation

significance levels. A significance level of 0·1 indicates a good correlation and a further decrease in the level indicates a higher correlation. During the period the stratopause height varied between 41-51 km, the temperature between 256–284° K and density between 0·81–3·45 gm/meter^3. The corresponding variation in the sunspot number was between 0-140 and the solar flux was between 72–176 units.

The above correlation coefficients show a good positive correlation between stratopause temperature and solar parameters, this is clearly brought out in Fig. 2. The absence of such correlation with neutral density is interesting as this is a function of temperature. The high correlation is, evidently because of the fact that the solar ultra-violet radiation of the range 2000–3000 Å is mainly absorbed by a minor constituent, ozone which is responsible for the large temperature in the entire stratosphere with a peak at the stratopause[4]. The same reason holds good for the higher daytime temperatures as compared to that of the nighttime (Fig. 1). Another observation that could be made from Fig. 1 is that the solar effect may be almost simultaneous, which results in large stratopause temperature around noon. The results of the earlier workers for whitesands[3] closely agree with our observations of higher daytime temperatures.

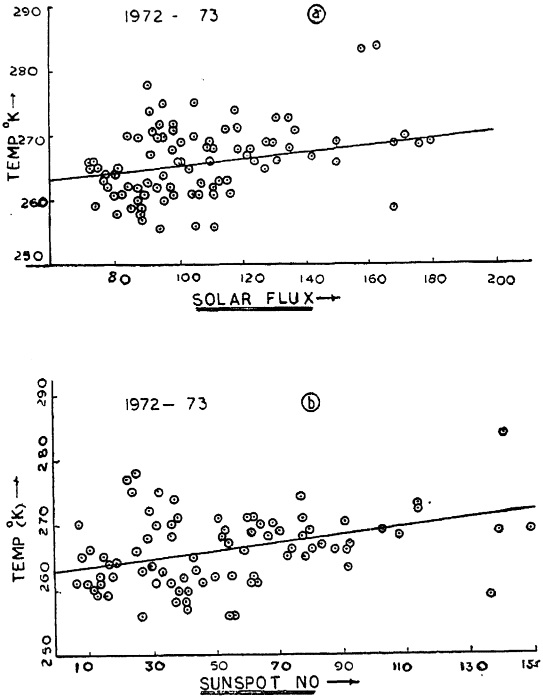

FIG. 2. Variations of temperature with (*a*) Solar Flux, (10·7 cm, Ottawa) and (*b*) Zurich Sunspot Numbers.

The observations that are made above, relating to the diurnal variations and the solar influence, will be confirmed by studying the daytime flights spread over a larger period. A detailed study taking into consideration the seasonal variations will be published elsewhere.

From the above studies the following conclusions are made. Higher daytime temperatures are observed compared to nighttime in the altitude region of 35–50 km for the winter atmosphere of Thumba. A good positive correlation is observed between the stratopause temperature and the solar parameters, and a poor correlation with the neutral density for, one biennial cycle, 1972–73.

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Waltair, *March* 15, 1976.

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1. Beyers, N. J. and Miers, B. T., *J. Atmos, Sci.*, 1965, **22**, 262.
2. —, — and Reed, R. J., *Ibid.*, 1966, **23**, 325.
3. Theon, J. S., Nordverg, W., Katchen, L. B. and Horvath, J. J., *Ibid.*, 1967, **24**, 428.
4. Schewentek, H., *J. Atmos., Terr. Phys.*, 1971, **33**, 1839.
5. Ramakrishna, S. and Seshamani, R., *Ibid.*, 1973, **35**, 1631.
6. Gaigerov, S. S., Kalikhman, M. YA., Sedov, V. E., Shvidkovsky, E. G., Tarasenko, D. A. and Zaichikov, B. P., *Space Research*, 1971, **11**, 799.
7. Fisher, R. A., *Statistical Methods for Research Workers* (Oliver and Boyd), p. 209.

THE CRYSTAL STRUCTURE OF *n-p*-CYANOBENZILIDENE-*p-n*-OCTYLOXYANILINE

As part of programme of systematic X-ray investigations of mesogenic compounds[1], the crystal structure of *n-p*-cyanobenzilidne *p-n*-octyloxyaniline (CBOOA) has been analysed in its stable solid phase. The compound is known to exhibit the following liquid crystalline phases on heating.

$$\begin{tikzcd} \text{Solid} \arrow[r, "73^\circ\text{ C}"] & \text{Smectic A} \arrow[r, "82{\cdot}5^\circ\text{ C}"] & \text{Nematic} \arrow[d, "106{\cdot}9^\circ\text{ C}"] \\ & & \text{Isotropic} \end{tikzcd}$$

Large, rectangular prismatic, yellow crystals of the compound were obtained by slow evaporation from a solution in *n*-heptane. Unit cell dimensions and space group were determined from conventional oscillation and Weissenberg photographs and are listed in Table I along with other relevant crystal data.

Three-dimensional intensity data were collected on multiple Weissenberg films using copper K_α radiation. The intensities were estimated visually and were corrected for Lorentz and polarisation factors, absorption and spot-shape effects.

TABLE I

Chemical formula	$C_{22}H_{26}N_2O$
Molecular weight	334·45
	$a = 15{\cdot}99 \pm 0{\cdot}04$ Å
	$b = 15{\cdot}91 \pm 0{\cdot}02$
	$c = 7{\cdot}66 \pm 0{\cdot}02$
	$\beta = 91 \pm 1°$
Space group	$p2_1/c$
	$Z = 4$
	$\rho_c = 1{\cdot}14$ gm . cm^{-3}
	$\rho_m = 1{\cdot}14$ gm . cm^{-3}

(Flotation in aqueous KI solution)
μ for copper K_α radiation = 5·61 cm^{-1}.

The structure was solved by the direct methods followed by Fourier synthesis. The positional as well as the isotropic thermal parameters of all non-hydrogen atoms were refined to R = 0·145 by block-diagonal structure-factor least-squares procedure using 1654 observed reflections. Further refinement of the structure is in progress.

Figure 1 gives the projection of the structure in the *bc*-plane. The molecules associate in pairs about centres of inversion, the two molecules in each pair being anti-parallel to each other. Along the *a*- and *c*-directions, adjacent pairs are related by cell translation and along the *b*-direction they are related by the 2_1 screw axes and the glide planes. Results of further refinement and the detailed description of the structural features will be discussed elsewhere.

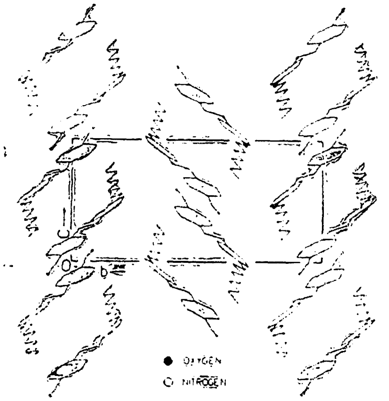

FIG. 1. Projection of the crystal structure in the *bc*-plane.

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1. Kalyani Vijayan and Vani, G. V., *Pramāṇa Supplement,* 1975, 1, 75.

K-SHELL PHOTOELECTRIC CROSS-SECTIONS AT 32·879 keV IN INTERMEDIATE Z-ELEMENTS

IN order to supplement the existing data on K-shell photoelectric cross-sections, measurements at 32·879 keV in elements Y, Zr, MO, Ag, Cd and Sn have been made. The method used was similar to the one reported earlier[1]. Photons of energy 32·879 keV were obtained from the K-conversion of 662 keV level in Ba^{137m}. The results obtained are compared with the available theoretical calculations[2-4] (Table I) and are found to agree with the theoretical calculations within the experimental uncertainties. The present results prove that the use of the theoretical values of the K-shell photoelectric cross-sections in our earlier determination[5] of the L-shell cross-section at 32·879 keV from the comparison of the intensity of L-shell fluorescent X-rays

TABLE I

Comparison of experimental data with theoretical calculations

z	Element	K-shell photoelectric cross-sections at 32·879 keV	
		Present measurements	Theoretical values
39	Y	2200±190	(*a*) 2300
40	Zr	2700±230	(*a*) 2450
42	Mo	2600±220	(*a*) 2900 (*b*) 2900
47	Ag	4600±390	(*a*) 4400 (*b*) 4350
48	Cd	4600±390	(*a*) 4550
50	Sn	5300±450	(*a*) 5350 (*b*) 5300 (*c*) 5300

(*a*)—Scofield; (*b*)—Schmickley-Pratt;
(*c*)—Rakavy-Ron.

with that of the K-shell fluorescent X-rays is justified.

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1. Allawadhi, K. L. and Sood, B. S., *Phys. Rev.*, 1975, 11 A, 1928.
2. Rakavy, G. and Ron, A., *Ibid.*, 1967, **159**, 50.
3. Schmickley, R. D. and Pratt, R. H., *Lockhead Research Laboratories Report*, L.M.S.C. 5–10–67–11 NYO–3839–9.
4. Scofield, J. H., *Lawrence Livermore Laboratory Report*, 1973, UCRL 51326.
5. Allawadhi, K. L., Mohanjit Singh and Sood, B. S., *Curr. Sci.*, 1973, 42, 86.

OBSERVED CORRELATION BETWEEN COMPOSITION AND MOLECULAR WEIGHT OF AMINO ACIDS IN PROTEINS

THE composition of various proteins, in particular, the distribution of amino acids has been a topic of considerable interest. Early work[1,2] quoted in Meister[3] would appear to yield some general conclusions, although some of these need revision in the light of recent analysis (Dayhoff[4]) which is based on primary sequence analysis. The latter analysis has yielded the relative frequency of occurrence of the amino acids according to the classification of polar, apolar, heterocyclic (and aromatic) and ionisable types.

The randomness or otherwise of the occurrence of these amino acids has been treated statistically[5-7] and tables are available to assist in assessing whether or not their occurrence in a particular protein may be considered as random. Other statistical studies[8-16] are aimed at the analysis of the relative frequency of occurrence of residues in relation to the characteristic secondary structure of the protein chain, with a view to using them in algorithms for predicting protein structure from the given sequence. So also analysis from the point of view of volume occupied by side chain is also available[17]. However as far as we are aware of, no study seems to have been made on the relative abundance of the amino acids in relation to the molecular weight of the constituents.

We have done this and the simple linear correlation that has been observed, is felt to be sufficiently interesting to merit reporting here. Though Dayhoff's Table D 23[4] includes 108 proteins, a few fibrous proteins and also some small peptides have been included in it. In view of this we have selected 32 proteins whose structure by x-ray crystallographic methods have been reported and hence the sequence is uniquely known. This we believe is a sufficiently good sample to yield statistically meaningful result. Figure 1 shows the relation between relative frequency of occurrence and the molecular weight.

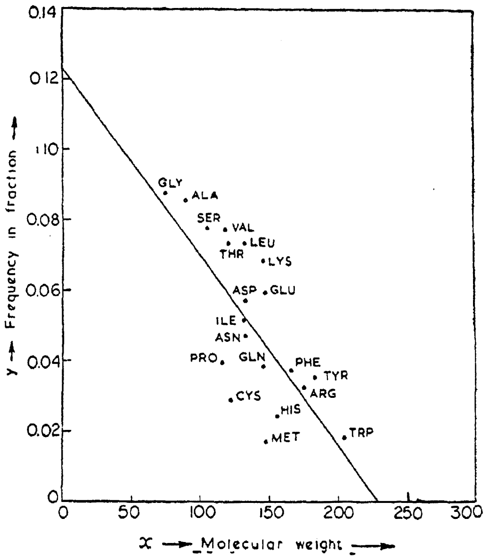

FIG. 1. Plot of molecular weight *vs.* the relative frequency of occurrence of amino acids in 32 proteins.

It is interesting to note that there is an approximately linear relation. The largest deviation from the above line is noticed for methionine and cystine and it is interesting that these two are amino acids containing sulphur. Among the rest, leucine (non-polar), lysine (polar) and histidine (heterocyclic) show somewhat large deviations.

The least squares equation to the line in Fig. 1 :

$$y = -0\cdot 00054\, x + 0\cdot 12445.$$

The equation leads to the result that nearly 67% of the amino acids are those having molecular weights less than 137 which is the mean of the 20 amino acids. At the two ends, the relative frequencies work out to be 8·4% for glycine and 1·4% for tryptophan. These are respectively about 70% more and 70% less than the mean (5%) to be expected if all the 20 amino acids are randomly distributed with equal probability.

Further analysis from this angle is in progress and results will be reported in due course.

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1. Tristram, G. R., *The Proteins* (Eds. Neurath, H. and Bailey, K., Academic Press, London, 1953), Vol. 1, Part A, p. 181.
2. —, *Advances in Protein Chemistry*, 1963, 18, 227.
3. Meister, A., *Biochemistry of Amino Acids*, Academic Press, New York, London, 1965, Vol. 1, Chap. I.
4. Dayhoff, M. O. and Hunt, L. T., *Atlas of Protien Sequence and Structure*, National Biomedical Research Foundation, Maryland, 1972, Table D 23, D–356.
5. Holmquist, R., *J. Mol. Evol.*. 1975, 4 (4).
6. Laird, M. and Holmquist, R., *Ibid.*, 1975, 4 (4), 261.
7. Black, J. A., Stenzel, P. and Harkins, R. N., *J. Theor. Biol.*, 1975, 50 (1), 161.
8. Chou, P. Y. and Fasman, G. D., *J. Mol. Biol.*, 1973, 74, 263.
9. — and —, *Biochemistry*, 1974, 13, 211.
10. — and —, *Ibid.*, 1974, 13, 222.
11. Burges, A. W., Ponnuswamy, P. K. and Scheraga, H. A., *Israel J. Chem.*, 1974, 12, 239.
12. Lim, V. I., *J. Mol. Biol.*, 1974, 88, 857.
13. —, *Ibid.*, 1974, 88, 873.
14. Nagano, K., *Ibid.*, 1973, 75, 401.
15. —, *Ibid.*, 1974, 84, 337.
16. Ptitsyn, O. B., *Ibid.*, 1969, 42, 501.
17. Chothia, C., *Nature*, 1975, 254, 304.

THE EVALUATION OF THE ELECTRONIC TRANSITION MOMENT FOR A BAND SYSTEM OF DIATOMIC MOLECULE

IT has been shown that the electronic transition moment[1] plays a very important role in the calculation of vibrational transition probabilities of diatomic molecules. The relative vibrational transition probability[2] $P_{v'v''}$ may be written as

$$P_{v'v''} = R_e^2 (\bar{r}_{v'v''}) q_{v'v''} \tag{1}$$

where $R_e (\bar{r}_{v'v''})$ is the electronic transition moment, $\bar{r}_{v'v''}$ is the *r*-centroid and $q_{v'v''}$ is the Franck-Condon factor of the band in question.

The intensity in emission $I_{v'v''}$ of a band may be written as

$$I_{v'v''} = KN_{v'}E^4_{v'v''}P_{v'v''} \tag{2}$$

in which K incorporates all the constants, $N_{v'}$ is the population of the level *v'*, $E_{v'v''}$ is the energy quantum of the band. Thus an array of relative $P_{v'v''}$ may be determined from $I_{v'v''}N_{v'}E^4_{v'v''}$ when the band intensities have been measured. From equations (1) and (2).

$$I_{v'v''} = KN_{v'}E^4_{v'v''}R_e^2 (\bar{r}_{v'v''}) q_{v'v''} \tag{3}$$

This equation may be used to determine the form of $R_e(\bar{r})$ for a band system.

Equation (3) may be employed to determine the relative variation of $R_e(\bar{r})$ with $\bar{r}$ for the band system. Once a smooth curve has been obtained which shows the relative variation of $R_e(\bar{r})$ with $\bar{r}$, equation (1) may be employed to determine a smoothed array of $P_{v'v''}$. Values of $R_e(\bar{r}_{v'v''})$ are read from the curve at values of $\bar{r}_{v'v''}$ appropriate to each band. $R_e^2(\bar{r}_{v'v''}) q_{v'v''}$ is then a relative measure of $P_{v'v''}$ for each band. The array of $P_{v'v''}$ thereby obtained may be normalised such that $P_{00} = 1$.

In the paper of Mishra and Khanna[3], the variation of $R_e(\bar{r}_{v'v''})$ with internuclear separation has been worked out by the method outlined by Nicholls[4] and it is found that the curve shown in the figure is consistant with the relation $R_e(\bar{r}_{v'v''}) =$ const. exp. $(0 \cdot 205\ r)$ over the range $1 \cdot 20$ Å $< r < 1 \cdot 45$ Å. The calculated $P_{v'v''}$ values for the bands of the A–X system of BF molecule are in fair agreement with the observed intensity distribution. Values of $P_{v'v''}$ are in good agreement with the values entered in the last column of Table I which are nothing but the product of F–C factor and *r*-centroir of the band. On this basis it can be inferred that vibrational transition probabilities for a band system of molecules for which $q_{00}\,\bar{r}_{00} \cong 1$, can be obtained without measuring the intensities of the bands. The details of this method along with the results of other band systems will be published elsewhere.

TABLE I

Data for BF (A–X) band system

Band	$_{v'v''}$ (Å)	$p_{v'v''}$*	$p_{v'v''}$
(0, 0)	1957·4	1·000	0·999
(0, 1)	2001·5	0·271	0·280
(0, 2)	2068·1	0·023	0·024
(1, 0)	1911·0	0·227	0·218
(1, 1)	1962·7	0·577	0·578
(1, 2)	2016·3	0·481	0·502
(1, 3)	2071·6	0·022	0·023
(2, 1)	1917·1	0·330	0·321
(2, 2)	1968·2	0·198	0·196
(2·3)	2021·3	0·539	0·558

* Data from Mishra and Khanna[3].

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---

1. Herzberg, G., *Molecular Spectra and Molecular Structure : Spectra of Diatomic Molecules* (Second Edn.), (D. Van Nostrand, New York, 1950.
2. Fraser, P. A., *Can. J. Phys.*, 1954, **32**, 515.
3. Mishra, P. K. and Khanna, B. N., *J. Quant. Spectrosc. Radiat. Transfer*, 1970, **10**, 703.
4. Nicholls, R. W., *Can. J. Phys.*, 1954, **32**, 722.

ADDUCTS OF BIS (ETHYLACETOACETATO) COPPER (II) WITH NITROGEN LIGANDS

Bonding[1-3] of additional ligands along the axis normal to the molecular plane of planar complexes like beta-diketonates and related compounds, which involve keto-enol tautomerism, have invoked a lot of interest in recent years. Since beta-keto esters are closely related to the beta-diketones, it was thought worthwhile to extend the investigation to such chelates and this communication reports some base adducts of bis-(ethyl acetoacetato copper (II) with substituted pyridines, *viz.*, –2, 3– and 4-methyl pyridines.

Ethanolic solution of cupric chloride was reacted with ethyl acetoacetate in (1 : 2) proportion, followed by dropwise addition of ammonia with constant stirring. The green product obtained was filtered off, washed with ethanol and ether and dried in a vacuum desicctor. Metal chelate and the nitrogen ligands (in 1 : 2 ratio) were taken in minimum quantity of ethanol and refluxed for thirty minutes. On cooling the solution overnight, crystalline compounds separated out.

Metal ion in the compounds was estimated by EDTA procedure. Conductance was measured in M/1000 acetone solution using a 'Toshniwal' conductance bridge. Magnetic susceptibility was measured on the solid specimen by Gouy method. Infrared spectra were recorded in nujol mulls with Unicam SP 200 Spectrophotometer. U.V. and visible spectra were recorded using M/100 chloro-from-base solution in Unicam SP 500 Spectrophotometer. The relevant analytical, conductance and magnetic susceptibility data are recorded in Table I.

Results and Discussion

The green crystalline compounds have the composition $Cu(EAA)_2L_2$ where 'L' is substituted pyridine. The compounds are stable in air, have low melting points. Acetone solutions of the complexes have low molar conductance values, ΛM being in the range of 2–4 mhos, indicating non-electrolytic nature.

Most of the absorption bands due to the free nitrogen ligands are shifted in the adducts indicating bonding to the metal, information being obtained by the "Finger Print" technique. ν (C=O) and ν (C=C) of the ester which occur at 1650 and 1630 cm^{-1} respectively shift to lower frequency region (1600 and 1525 cm^{-1}) on chelation and again shift to higher frequency (1620 and 1540 cm^{-1} respectively) in the nitrogen base adducts providing additional evidence for bonding of the ester and nitrogen ligand. Direct evidence could not be obtained as ν (Cu–O) and ν (Cu–N) were beyond the range of instrument used.

An intense band appeared in the U.V. region around 260 nm ($\epsilon = 15000$) in solution spectrum of the chelate and base adducts due to the more probable $\pi \rightarrow \pi^*$ transition rather than charge transfer. In the visible region, a broad absorption band was observed around 660 nm ($\epsilon = 60$) in the chelate which was found around 650 nm ($\epsilon = 85$) in the base adducts. On coordination of the nitrogen ligands there is not much shift in the position of the absorption band though the intensity increases.

Table I

Analysis, M.P., Conductance and magnetic susceptibility data of bis-(ethyl acetoacetato) Copper(II) complexes

Compounds	Colour and form	MP (° C)	ΛM ohm^{-1} cm^2	μ_{eff} (B.M.)	% Metal		% N	
					Reqd.	Found	Reqd.	Found
$Cu (EAA)_2 (\alpha\text{-pic})_2$	Green crystalline	160	2·8	1·76	12·56	12·0	5·52	5·3
$Cu (EAA)_2 (\beta\text{-pic})_2$	do.	180	3·0	1·74	12·56	12·1	5·52	5·25
$Cu (EAA)_2 (\gamma\text{-pic})_2$	do.	175	4·2	1·78	12·56	11·9	5·52	5·32

This is in conformity with some earlier observations[4]. Hence the complexes are six-coordinated, presumably having a tetragonally distorted octahedral configuration. Though distorted octahedral Cu (II) complexes have three transitions ($2B_{1g} \rightarrow 2B_{2g}$ $2B_{1g} \rightarrow 2E_1$, $2B_{1g} \rightarrow 2A_{1g}$), often the three bands appear[5] in a single broad asymmetric band envolope. The base adducts appear to be far more stable than their beta-diketonato analogues since in absence of the added base, there is no change in the position of absorption band in solution spectrum indicating "no appreciable dissociation" of the ligands.

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1. Graddon, D. P., *Nature* (London), 1959, 183, 1610.
2. Walker, W. R., *Aust. J. Chem.*, 1961, 14, 161.
3. Gillard, R. D. and Wilkinson, G., *J. Chem. Soc.*, 1963, p. 5399.
4. Graddon, D. P. and Mockler, G. M., *Aust. J. Chem.*, 1968, 21, 907.
5. Agamber, C. A. and Orrel, K. G., *J. Chem. Soc.* (A), 1969, p. 897.

BROMINATION OF ACETANILIDE CATALYSED BY ANHYDROUS FERRIC CHLORIDE, ANHYDROUS ZINC CHLORIDE AND IODINE BROMIDE

It is a common practice in synthetic organic chemistry to speed up the halogenation of aromatic compounds by the addition of halogen carriers such as iodine, iron powder, anhydrous halides of iron, zinc and aluminium, silver salts and pyridine[1,2]. Iodine bromide has been shown to be the effective catalyst in the presence of added amounts of iodine in the bromination of aromatic compounds[3-5].

In the presence of IBr, $ZnCl_2$ and NaCl as catalysts, the bromination of anisole in 85% aqueous acetic acid has been shown[6] to proceed by simultaneous catalysed and uncatalysed processes. In the present investigation[7], the bromination of acetanilide in the presence of added salts was followed kinetically[3] at 30° C by mixing 5 ml each of 0·02 M solutions of acetanilide and bromine in anhydrous acetic acid, allowing them to react for definite intervals of time and arresting the reaction by the addition of 10 ml of an aqueous solution of 1·0 g of KI. In the $FeCl_3$-catalysed reaction, the KI solution also contained 0·33 g of NaF, which complexes with Fe^{3+} preventing the oxidation of KI by Fe^{3+}.

In the absence of catalysts, the acetanilide-bromine reaction was found to be of the third order in the range 0·02–0·01 M. The individual orders determined by the isolation method[3] were 1·0 for acetanilide and 1·9 for bromine, in agreement with recent work[8]. When anhydrous ferric chloride was added to the reaction mixture, there was an increase in the rate of the reaction. When the concentration of $FeCl_3$ in the reaction mixture was increased ($3 \cdot 6 \times 10^{-4}$ to 54×10^{-4} M), there was a steady increase in the observed average third order rate constant for 20–50% bromine consumption. A similar catalysis was observed in the presence of added anhydrous $ZnCl_2$ and iodine (which forms iodine bromide *in situ*), as seen from Table I.

The rate of bromination of acetanilide in the presence of added salts (A) may be expressed as follows:

$$-\frac{d(Br_2)}{dt} = k(ArH)(Br_2)^2 + k_c(ArH)(Br_2)^2(A)$$

or (1)

$$k_3 = k + k_e(A) \qquad (2)$$

where k_3 is the observed third order rate constant, and k and k_c are the rate constants for the uncata-

Table I

Effect of added salts on the bromination of acetanilide

Acetanilide = 0·010 M Solvent: Anhy. HOAc
Bromine = 0·010 M Temp. 30·0 ± 0·1° C

$FeCl_3$ × 10^4 (M)	$k_3 \times 10^{-2}$ (lit²/mole²/min)	$ZnCl_2$ × 10^4 (M)	$k_3 \times 10^{-2}$ (lit²/mole²/min)	IBr × 10^4 (M)	$k_3 \times 10^{-2}$ (lit²/mole²/min)
3·6	3·73±0·03	2·6	3·80±0·04	0·8	3·78±0·06
15·0	4·07±0·04	5·3	4·07±0·02	1·6	4·05±0·10
32·7	4·53±0·06	8·4	4·45±0·03	2·9	4·55±0·20
54·0	5·00±0·05	16·6	5·34±0·02	4·8	5·14±0·38

All the concentrations quoted are those after mixing.

lysed and catalysed reactions, respectively. A plot of k_3 *versus* the molar concentration of A was found to be linear in each case. From the slopes and intercepts of these lines k and k, respectively, were calculated. The values thus obtained are given in Table II.

TABLE II

Relative reactivity of the added salts in the bromination of acetanilide

Added salt	Added salt conc. range $\times 10^4$ (M)	$k \times 10^{-2}$ (lit^2/mole^2/min)	$k_c \times 10^{-4}$ (lit^3/mole^3/min)	Relative reactivity
Nil	..	3·5	..	..
$FeCl_3$	3·6–54·0	3·7	2·5	1·0
$ZnCl_2$	2·6–16·6	3·5	11·2	4·5
IBr	0·8–4·8	3·5	35·2	14·1

From the last column of Table II it is seen that the order of reactivity of the three halogen carriers is $IBr > ZnCl_2 > FeCl_3$. The role[9] of these catalysts is to polarise the bromine molecule and to remove Br^- as ABr^- in the rate-determining step. Their relative reactivity may be attributed to the following order of stability of ABr^-:

$$IBr_2^- > ZnCl_2Br^- > FeCl_3Br^-.$$

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---

1. de la Mare, P. B. D. and Ridd, J. H., *Aromatic Substitution: Nitration and Halogenation*, Butterworths, London, 1959, p. 105.
2. Braendlin, H. P. and McBee, E. T., In *Friedel-Crafts and Related Reactions*, Ed. Olah, G. A., Interscience Publishers, New York, 1964, Vol. 3, Part II, p. 1530.
3. Yeddanapalli, L. M. and Gnanapragasam, N. S., *J. Chem. Soc.*, 1956, p. 4934.
4. Gnanapragasam, N. S., Venkateswara Rao, N. and Yeddanapalli, L. M., *J. Ind. Chem. Soc.*, 1959, 36, 777.
5. Kothandaraman, G., Rajaram, J. and Kuriacose, J. C., *Proc. Ind. Acad. Sci.*, 1970, 72 A, 144.
6. Gnanapragasam, N. S. and Joseph, N., *Curr. Sci.*, 1971, 40, 83.
7. Ramanujam, R., *M.Sc. Project Report*, University of Madras, April 1976.
8. Nanian, M. J. and Ganesan, R., *Z. Physik. Chem. Neue Folge*, 1974, 91, 183.
9. Joseph, N. and Gnanapragasam, N. S., *Curr. Sci.*, 1972, 41, 288.

CHROMATOGRAPHIC IDENTIFICATION OF CERTAIN SULFOXIDES OF CYSTEINE PRESENT IN ONION (*ALLIUM CEPA* LINN.) EXTRACT

Introduction

VIRTANEN AND MATIKKALA[1,2] isolated three sulfoxide amino acids, *viz.*, cycloalliin, S-methyl cysteine sulfoxide and S-propyl cysteine sulfoxide from methanolic extracts of onions (*Allium cepa* Linn.). Bandyopadhyay *et al.*[3] reported that some of the sulphur compounds of the etherial extracts of onion possess carbonyl and hydroxyl groups, but they did not identify them. The therapeutic effects of these extracts[4] created some interest in their chemistry. In a preliminary study the diethyl ether extract of onion showed the presence of amino acids. This posed the question whether the suspected carbonyl compounds reported to be present in the etherial extract of onion[3] really belonged to some of the sulfoxide amino acids of onion. In order to study this question the sulfoxide amino acids were separated from etherial and ethanolic extracts of onion, identified and compared by paper chromatography.

Experimental

Diethyl ether extract of fresh onion was prepared[3] and petroleum ether (B.P. 40–60) soluble fraction[4] was removed from it as reported previously. The insoluble fraction was further extracted with 20 ml of 1 NHCl and centrifuged. This process was repeated thrice. The combined solution was neutralised to litmus with 1 N NaOH and acetone was added to make it 50% acetone. The crystals separated on cooling were recrystallised from 50% acetone-water[1]. The crystals were analysed for amino acids by paper chromatography using butanol : acetic acid : water system. A concentrated ethanolic extract of onion was prepared and the sulfoxide amino acids were separated by column chromatography according to the method of Carson and Wong[5] with certain modifications that IR–120 (H^+ form)[1] and IRA–93 (OH) were used in place of Dowex 50 – X_4 (H^+ form) and Duolite A–4 (amino form) respectively. To study the total amino acid pattern a sample of crystals obtained from the effluent from IR–120 were analysed by both one and two dimensional chromatography. The amino acids absorbed finally on IRA–93 (OH) were eluted with 0·1 N formic acid and one litre fractions were evaporated, acidified and further concentrated *in vacuo*[5]. Crystals separated were analysed by two-dimensional paper chromatography using butanol : acetic acid : water system first (12 : 3 : 5) and phenol ammonia : water system second[6]. Ninhydrin was used as the colour developing reagent. Each fraction was further recrystallised from 2 N HCl and analysed again by descending chromatography (Bu : Ac : H_2O). Standard S-methyl and S-propyl cysteine sulfoxides and cycloalliin were used as reference compounds.

Results and Discussion

The amino acids obtained from the ether extract (yield 0·1 g/Kg onion) on descending chromatography gave spots corresponding to S-methyl cysteine sulfoxide ($R_f = 0{\cdot}17$) and S-propyl cysteine sulfoxide ($R_f = 0{\cdot}35$). A third spot also appeared ($R_f = 0{\cdot}63$) which may be S-propyl cysteine[1]. The amino acids obtained from the ethanolic extract (yield 0·65 g/Kg onion) on chromatography gave spots corresponding to the above two sulfoxide amino acids and also cycloallin ($R_f = 0{\cdot}24$). The identity of the three sulfoxide amino acids in the above preparation was further proved by two-dimensional chromatography. About twelve unidentified amino acids also appeared on the same chromatogram. They may be the other onion amino acids reported in a similar study[7]. By recrystallisation a mixture of the above three sulfoxide amino acids was obtained from fraction 1 [eluent from IRA–93 (OH) yield 0·3 g/Kg onion] and pure cycloalliin from fractions 2 and 3. yield 0·2 g/Kg onion.

Some of the onion sulfur compounds which, Bandyopadhyay *et al.*[3] separated from the ether extract may therefore belong to S-methyl and S-propyl cysteine sulfoxides and their degradation products reported elsewhere[8]. The therapeutic effects of the ether extract of onion and its sulphur compounds[4,9–11] and also the importance of its sulfoxide amino acids with relation to sulphur metabolism[12] warrant further investigation on this vegetable.

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---

1. Virtanen, A. I. and Matikkala. E. J., *Acta Chem. Scand.*, 1959, 13, 1898.
2. Matikkala. E. J. and Virtanen, A. I., *Suomen Kemistilehti*, 1957. 30 B, 219.
3. Bandyopadhyay, C., Srirangarajan, A. N. and Sreenivasan, A., *J. Chromatog.*, 1970, 47, 400.
4. Mathew. P. T. and Augusti. K. T., *Ind. Jour. Exptl. Biol.*, 1973. 11, 573.
5. Carson, J. F. and Wong, F. F., *Jour. Org. Chem.*, 1961, 26. 4997.
6. Smith, I., In Ivor Smith (editor), *Chromatographic and Electrophoretic Techniques*, Vol. I. *Chromatography*, Williams Heinemann Medical Book Ltd., London, 2nd ed., 1960, Chapter 5, p. 82.
7. Matikkala, E. J. and Virtanen, A. I., *Acta Chem. Scand.*, 1967, 21, 2891.
8. Johnson, A. E., Nursten, H. E. and Williams, A. A., *Chem. and Ind.*, 1971, 1, 556.
9. Augusti, K. T., Benaim, M. E., Dewar, H. A. and Virden, R., *Atherosclerosis*, 1975, 21, 409.
10. — and —, *Clin. Chim. Acta*, 1975, 60, 121.
11. —, *Ind. Jour. Exptl. Biol.*, 1976, 14, 110.
12. Maw, G. A., In A. Senning (editor), *Sulphur in Organic and Inorganic Chemistry*. Vol. 2, Marcel Dekkar, Inc., New York, 1st Edn., 1972, Chapter 1, p. 115.

CHEMISTRY OF *TERMINALIA* SPECIES

Part XV*. Chemical Examination of *T. procera* Roxb.

IN the course of our studies on the heartwoods of *Terminalia* species[1–4], a sample of heartwood of *T. procera* Roxb. was secured from Andaman islands. Preliminary examination showed the presence of tannins asd phenolic compounds, but no triterpenes.

The hexane extract contained β-sitosterol (m.p. 136–37°, acetate m.p. 125–26°), identified by comparison with an authentic specimen. The chloroform solubles contained three components which could be separated by column chromatography on silica gel. From the analysis and by comparison with the authentic samples, they were identified as (*i*) ellagic acid [m.p. 360°, blood red colouration with Greismeyes's reagent[5], ν_{max}^{Nujol}: 3570(m), 3490, 1730–1705 (br), 1625, 1585, 1540 cm^{-1}, λ_{max}^{EtOH}: 255, 366, with NaOAc 256, 278, 355[6]] (*ii*) 3, 3′-di-O-methyl ellagic acid [yield 0·03%, m.p. 325–27°, blood red colouration with Greismeyer's reagent[5], ν_{max}^{Nujol}: 3460 (br), 1715, 1580, 1550 cm^{-1}, λ_{max}^{EtOH}: 248 (log ϵ 4·75), 372 (log ϵ 4·15) unaffected with NaOAc or $AlCl_3$[6]] and (*iii*) 3, 3′, 4-tri-O-methyl ellagic acid [yield 0·035%, m.p. 285–87°, green-brown-yellow colouration with Greismeyer's reagent[5], ν_{max}^{Nujol}: 3440 (br), 1720, 1580, 1550 cm^{-1}, λ_{max}^{EtOH}: 249 (log ϵ 4·65), 370 (log ϵ 4·05) unaffected with NaOAc or $AlCl_3$[6]]. Their identification was further confirmed by the preparation of tetra-O-methyl ellagic acid (m.p. 340–42°) using K_2CO_3 and dimethyl sulphate in acetone. Ellagic acid was obtained (yield 0·5%) from the acetone extracts of the heartwood.

Ellagic acid is a common constituent of a number of species of *Terminalia*. The 3, 3′-di-O-methyl ellagic acid was obtained from *Euphorbia formosanum* Hey[7] and it occurs as a glucoside in the heartwood of *T. paniculata* Roth[8]. The 3, 3′, 4-tri-O-methyl ellagic acid was isolated from the bark and heartwood of *Eugenia maire* A. Cunn[9]. The co-occurrence of these two methyl ethers has been found for the first time in *T. procera*.

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* Part XIV: Ramachandra Row, L. and Suryanarayana Murthy, P., *Ind. J. Chem.*, 1970, 8, 1047.

1. Pearson, R. S. and Brown, F. P., *Commercial Timbers of India*, Vol. I, Government of India, Central Publications, Calcutta, 1932.
2. Ramachandra Row, L. and Ramakrishnam Raju, R. *Tetrahedron*, 1967, 23, 879; *Curr. Sci.*, 1965, 34, 177.
3. — and Subba Rao, G. S. R., *Tetrahedron*, 1962, 18, 827.
4. —, Suryanarayana Murty, P., Subba Rao, G. S. R., Sastry, C. S. P. and Rao, K. V. J., *Ind. J. Chem.*, 1970, 8, 772.
5. Lederer, E., *J. Chem. Soc.*, 1949, p. 2115.
6. Jurd, L., *Chem. and Ind.*, 1959, p. 261; *J. Am. Chem. Soc.*, 1959, 81, 4610.
7. Shinoda, H. and Kun, C. P., *J. Pharm. Soc.*, Japan, 1931, 51, 50.
8. Ramachandra Row, L. and Subba Rao, G. S. R., *Tetrahedron*, 1962, 18, 357.
9. Briggs, L. H., Cambie, R. C., Lowry, J. B. and Seelye, R. N., *J. Chem. Soc.*, 1961, p. 642.

THE UTILITY OF MANGANESE IN LACTOSE MEDIUM TO DIFFERENTIATE RHIZOBIA FROM AGROBACTERIA

AGROBACTERIA are the most common bacterial contaminants which resemble rhizobia and are often confused with them during the routine isolation procedures. However, certain biochemical tests[1,2] have been proposed earlier to differentiate rhizobia from many species of *Agrobacterium*. In this connection, Clark[3] has reported that a modified Bergersen's medium[4] containing lactose and 20 m.e. Mn^{2+}/1 could preferentially support the growth of all the species of *Agrobacterium* but exclude strains of clover and medic-rhizobia. In the present investigation, this finding has been verified extensively with tropical rhizobia of diverse origin comprising 115 isolates from *Cicer arietinum*, 12 from *Sesbania bispinosa* and *S. sesban*, and 8 isolates of *Agrobacterium* sp.; the latter was isolated as contaminants during routine isolation of nodule bacteria. All the isolates were inoculated on agar slopes of the following composition: lactose, 5·0 g; KNO_3, 1·0 g; $MgSO_4.7H_2O$, 0·1 g; Na_2HPO_4 (anhydrous), 0·18 g; agar 12·0 g; FeEDTA (0·25% W/v), 10·0 ml; $MnSO_4$ (33·5% W/v), 10·0 ml; pH, 6·8 and volume made upto 1000 ml by distilled H_2O. After incubating the cultures for 14 days, at 28° ± 1° C, observations were recorded for visible growth. Infectivity of all the isolates was tested on their homologous hosts according to the method of Wieringa and Bakhuis[5].

It was interesting to note that all the isolates of rhizobia which nodulated their hosts could not also grow on the manganese lactose medium while the isolates of *Agrobacterium* showed luxuriant growth, thereby lending support to Clark's finding[3] and confirming the utility of the test as a diagnostic feature to differentiate *Rhizobium* from *Agrobacterium* (Table I).

TABLE I

Growth of rhixobia and agrobacteria on modified Bergersen's medium containing lactose and 20 m.e. Mn^{2+}/1

Strains	Isolated from	No. of strains found positive	
		Growth on lactose-manganese medium	Nodulation on homologous host
Cicer-rhizobia (115)	*Cicer arietinum*	..	115
Sesbania-rhizobia (10)	*Sesbania bispinosa*	..	10
Sesbania-rhizobia (2)	*S. sesban*	..	2
Agrobacterium (5)	*C. arietinum*	5	..
Agrobacterium (3)	*S. bispinosa*	3	..

Figures in parenthesis are the number of isolates.

Our sincere gratitude is due to Dr. N. S. Subba Rao, Head, Division of Microbiology, Indian Agricultural Research Institute, New Delhi, for his helpful discussion and critical comments.

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1. Kleczkowska, J., Nutman, P. S., Skinner, F. A. and Vincent, J. M., In *Identification Methods for Microbiologists*, Part B, Ed. Gibbs, B. M. and Shapton, D. A., Acad. Press, London, New York, 1968, p. 51.
2. Gaur, Y. D., Sen, A. N. and Subba Rao, N. S., *Curr. Sci.*, 1973, 42, 545.
3. Clark, A. G., *Appl. Bact.*, 1969, 32, 248.
4. Bergersen, F. J., *Aust. J. Biol. Sci.*, 1961, 14, 349.
5. Wieringa, K. T. and Bakhuis, J. A., *Plant and Soil.*, 1957, 8, 254.

ACETYLCHOLINESTERASE HETEROGENEITY IN THE BRAIN OF THE FROG *RANA CYANOPHLICTIS*

ACETYLCHOLINESTERASE (E.C. 3.1.1.7) has been found in the innervated tissues of all the vertebrates and invertebrates that have been studied[1]. The Significance of this enzyme has been demonstrated by the prolonged duration of ACh effects seen after administration of esterase inhibitors[2]. Baldwin and Hochachka[3] have studied the isoenzymes of AChE in the brain of rainbow trout, as a function of temperature acclimation. However no information regarding the isozyme pattern of acetylcholinesterase in the brain of frog is available. Since frog is the common experimental animal in biological research and in view of the importance of AChE in neuronal activity, it was felt desirable to initiate the present study. An attempt has been made here to resolve the different isozymal forms of the enzyme in the cortical region of the brain of frog.

Specimens of *R. cyanophlictis* of medium size (22–27 g) were maintained in the laboratory aquaria at 23 ± 2° C. The animals were fed on the leg muscle of frog once in three days. They were killed by decapitation and the brain was dissected and removed at 0° C. The cortex of the brain was isolated in amphibian Ringer[4] at 0° C and immediately used for analysis.

The isozymic pattern of acetylcholinesterase was studied by polyacrylamide gel electrophoresis (disc electrophoresis) developed by Davis and Ornstein[5,6]. The gel was formed from two organic monomers, acrylamide and bisacrylamide, the latter being the cross-linking agent. The catalyst used to initiate polymerization cross-linking reaction was tetramethylethylenediamine (TEMED).

The matrix was prepared in small glass tubes with three gel layers. The lower gel (consisting of acrylamide. bisacrylamide, potassium fericyanide riboflavin, TEMED and tris) was overlaid with 2 mm of distilled water and allowed to polymerize for 40 minutes under a fluorescent lamp after which the water layer was removed and gently washed with upper gel which consisted of tris, phosphoric acid, acrylamide, bisacrylamide and riboflavin. The upper gel was then laid similarly with 2 mm of distilled water and allowed to polymerize for 40 minutes after which the water layer was gently removed.

A 10% homogenate of the cortical matter of the brain of frog was prepared in phosphate buffer pH 7, and 0·1 ml of it was thoroughly mixed with 0·1 ml of upper gel solution and laid as the third gel layer and allowed to polymerize for 40 minutes. The tubes were run at 6 milliamps per tube (DC power supply model CM 07/02 Sr. No. 111, Toshniwal) for an hour in tris-glycine buffer at pH 9·5.

After the electrophoretic run, the tubes were removed and the gel was gently isolated from the glass tube. It was divided into two vertical halves. One half was further cut into 2·5 mm bits and processed for quantitative estimation of AChE activity by the colorimetric method of Hestrin[7]. The activity of AChE was expressed as micromoles of Ach hydrolysed per mg protein per minute. The other half of the gel was similarly processed for proteins by microbiuret method[8].

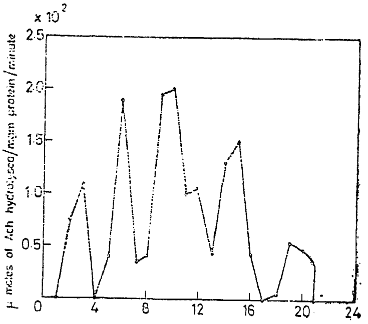

FIG. 1. Profile of acetylcholinesterase (AChE) in the cerebrum of frog. ' X ' axis represents section numbers of gel bits processed for the activity level of AChE. Peaks (starting from section No. 1) are designated as A_1, A_2, A_3, A_4 & A_5.

It is clear from Fig. 1 and Table I that by polyacrylamide gel electrophoresis, cortical acetylcholin-

TABLE I

Brain acetylcholinestrase heterogeneity in frog, Rana cyanophlictis

Body wt. (g)	Brain wt. (mg)	Isozymal forms				
		A_1	A_2	A_3	A_4	A_5
26·4 ± 3·1	80 ± 14	*110 ± 27	197 ± 29	202 ± 56	152 ± 20	55 ± 15
% change		58·8	97·5	100	75·2	27·2

* Activity expressed as micromoles of Ach hydrolysed/mg protein/minute. Each value represents mean ± SD of 2 observations.

For each observation brain (cortical matter) from 3 animals was pooled.

% change is calculated with reference to the activity level of peak A, which is considered 100.

esterase could be resolved into 5 isozymal forms designated as A_1, A_2, A_3, A_4 and A_5. All the five isozymes exhibited anodal migration (so distinguished because of the disc electrophoresis run in Tris-Glycine-buffer at pH 9·5 in which the flow of current was unidirectional, from cathode to anode). It is also clear (Fig. 1) that the second and the third isozymal forms (A_2 and A_3) exhibited comparatively higher activity. Of these, the form, designated A_3, exhibited the highest activity. Of the two comparatively fast moving forms, A_5 exhibited the least activity and the activity level of slowest moving isozyme (A_1) was approximately double that of A_5. Thus from the isozymic pattern of AChE in the cortical matter of the frog, *R. cyanophlictis*, it appears that in general the animal depends more on the slow moving anodal forms for its metabolic activity under normal physiological conditions, compared to the fast moving ones, since the activity of the fromer is higher.

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Bangalore 560 001, *March* 24, 1976.

1. Nachmansohn, D., *Handbook of Experimental Pharmacology*, (G. B. Koelle, ed.), Springer-Verlag, Berlin, 1963, **15**, 701.
2. Katz, B., The Sherrington Lectures as in *Handbook of Neurochemistry*, 4th ed. by A. Lajtha, 1969, p. 263.
3. Baldwin, J. and Hochachka, P. W., *Biochem. J.*, 1970, **116**, 883.
4. Cavanaugh, *Formulae and Methods* (Marine Biological Laboratory, Woods Hole, Mass., 1956).
5. Davis, B. J., *Ann. N.Y. Acad. Sci.*, 1964, **121**, 404.
6. Ornstein, L., *Ibid.*, 1964, **121**, 321.
7. Hestrin, S., *J. Biol. Chem.*, 1949, **180**, 249.
8. Itzhaki, R. T. and Glick D. M., *Analyt. Biochem.*, 1964, **9**, 401.

THE EFFECT OF "PAPER FACTOR" ON THE FECUNDITY AND HATCHABILITY IN *DYSDERCUS CINGULATUS* (HETEROPTERA: PYRRHOCORIDAE)

THE "Paper Factor" (PF), an inhibitor of embryonic development, was earlier reported to be effective only on *Pyrrhocoris apterus*[1]. Following the report of its morphogenetic effect on *Dysdercus koenigii*[2], the authors studied its ovicidal effect on *D. cingulatus*. The bugs were regularly reared in our laboratory at 28 ± 1° C, 70 ± 5% RH and 12 hour photoperiod. They were fed on soaked catton seeds. The 'PF' was extracted from paper using petroleum ether as solvent and after evaporation, the residue was redissolved in acetone. The female bugs were topically treated with 1 μl (30 μg) of 'PF'. Each pair was kept in a separate petridish. 150 pairs were used in this study. The eggs laid by each female and their hatchability were recorded. Acetone treated controls and normal untreated insects had no differences. Normal bugs laid 70 eggs on an average in each batch and almost all hatched. The experimental insects were treated with 'PF' at different stages of imaginal life. In group A, 32 freshly ecdysed females were treated. Of them 18 failed to mate and did not oviposit. Among those that mated and oviposited, the hatchability was 49% and 63% in the first two batches respectively. In group B, the 'PF' was applied to the females on third day after adult ecdysis; in group C, one day prior to first oviposition; and in group D, it was applied for three days continuously after first oviposition. The oviposition and percentage of hatchability is shown in Table I.

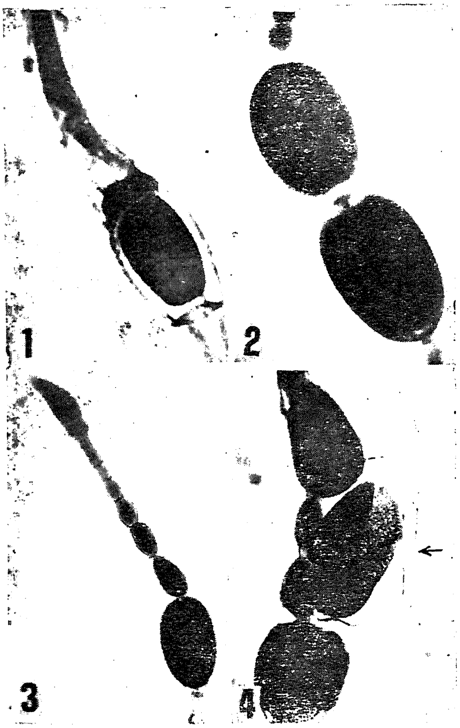
TABLE I
No. of eggs/percentage of hatchability

	A	B	C	D
I Batch	65/49	61/52	63/25	62/19
II Batch	67/63	52/77	51/69	51/6

Figures indicated are averages of minimum 20 insects except in A.

Irrespective of when the 'PF' was applied during the gonadal cycle, there has been a reduction in the number of eggs laid and in the number of larvae hatched. However, the mature eggs (C) appear to be more sensitive for the treatment and inhibition was found less in second batch except in group D, where the low rate in the second batch of eggs may be due to the large amount of active substance applied to the insect. Slama, *et al.*[3] stated that in *Pyrrhocoris apterus*, the treatment of normal females just after adult ecdysis with large doses of juvenoids had no profound influence on the course of its reproductive cycle and that they deposited perfect eggs. But in the present investigation, it has been observed that when freshly ecdysed 'females of *Dysdercus cingulatus* were treated with 'PF', some of the bugs did not even mate or oviposit and in those which mated and oviposited there has been a considerable inhibition.

When the treated bugs were dissected prior to oviposition, some ovarioles had only one developed egg, some had two, while in several others the ovaries were in a degenerated condition. In few cases when they were dissected after oviposition, some of the eggs were found attached to the ovarioles similar to those reported by Divakar and Novak[4] (Figs. 1–4).



FIGS. 1–4. Photomicrographs of the ovarioles affected by 'Paper Factor' in *Dysdercus cingulatus*. Fig. 1. Degenerated condition of the ovariole. Fig. 2. Development of only two eggs in the ovariole instead of six. Fig. 3. Development of only one egg in the ovariole. Fig. 4. Abnormality in the ovariole where a well-developed egg of the previous batch (marked with arrow) was found attached to the developing oocytes of the next batch.

In a separate experiment when a few freshly ecdysed males were treated and allowed to mate with normal untreated females of the same age, there has been a reduction in hatchability. This may probably be due to transfer of the active substance during copulation[5,6] or due to defective sperms.

Thus the present investigation conclusively shows that the "Paper Factor" has considerable effect in inhibiting the reproduction of the red cotton bug, *Dysdercus cingulatus*.

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---

1. Slama, K. and Williams, C. M., *Nature* (Lond.), 1966, 210, 329.
2. Saxena, K. N. and Williams, C. M., *Ibid.*, 1966, 210, 441.
3. Slama, K., Romanuk, M. and Sorm, F., *Insect Hormones and Bioanalogues,* Springer-Verlag, Wien, New York, 1974, p. 239.
4. Divakar, B. J. and Novak, V. J. A., *Int. Symp. Regulation of Insect Reproduction,* Liblice, Czechoslovakia, 1974.
5. Masner, P., Slama, K. and Landa, V., *Nature* (Lond.), 1968, 219, 395.
6. Judson, P., Divakar, B. J. and Kishen Rao, B., *Curr. Sci.*, 1976 (in press).

ANTAGONISTIC EFFECT OF THIAMINE ON INDOLEACETIC ACID IN GREEN GRAM (*PHASEOLUS RADIATUS* L.)

A VARIETY of compounds can inhibit growth in the presence of auxin and reduce the effectiveness of the auxin, but its mechanism is not clear. Evans and Ray[1] have shown that after treatment with IAA, corn and oat coleoptile segments continue to grow at a slow rate for some time. Suddenly, a substantial increase in the rate of elongation occurs. Some investigators feel that IAA acts as an allosteric[2]. No compound has yet been found which exerts strong competitive antagonistic action towards the plant growth hormone IAA. The present study has been designed to reveal certain facts with regard to this problem.

Seeds of green gram var. 525, were allowed to germinate and grow in petridishes for about 8 days in the dark. The vitamin B_1 (5 ppm) and IAA (5 ppm) treatment was given only for 24 hours, to avoid fungal or bacterial infection. The seedlings were then allowed to grow in distilled water in petridishes. Respiratory activity was measured by the usual Warburg manometric technique. The protein and soluble nitrogen were estimated using the method of Markham[3] and Thimann and Loos[4]. IAA at 5 ppm, although inhibitory at the initial stages, promoted the growth to about 15% over control. The stimulatory effect of B_1 (5 ppm) on growth (Fig. 1) gradually declined from about 14% to nearly 10% inhibition, over the control. The combined effect of IAA and B_1 was quite inhibitory. The effectiveness of auxin was reduced by thiamine in relation to growth (elongation). The respiratory activity (Table I) was inhibited by IAA. The stimulatory effect of IAA on growth in later stages of growth was not due to the increased respiratory activity. Thiamine (B_1) reversed the respiratory inhibition by IAA thus indicating the antagonistic behaviour.

With IAA treatment, there was a reduction in protein and soluble nitrogen contents. Thiamine (B_1) treatment caused an increase in protein nitrogen content when compared to that of control as well as with that of IAA treatment. The decrease in soluble nitrogen content caused by IAA treatment was counteracted or reversed by B_1 treatment particularly on the 4th day. On the sixth day IAA treatment showed an increase in soluble nitrogen with a concomitant decrease in protein nitrogen as compared to thiamine treatment. This apparently indicates that B_1 treatment caused an enhanced protein synthesis (compared to control and IAA) and that IAA treatment reduced the protein nitrogen while increasing the soluble nitrogen content. Thus, the antagonistic effect of B_1 and IAA was partly revealed from the behaviour of protein metabolism.

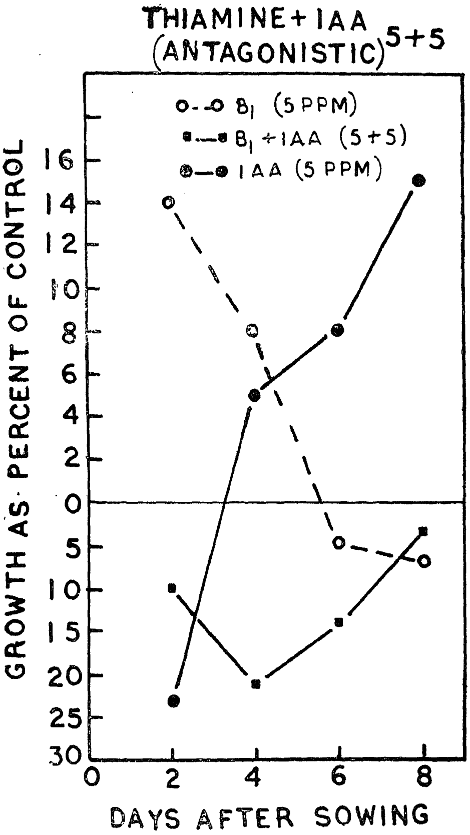

FIG. 1

Thiamine reversed the respiratory inhibition by IAA and the decrease in soluble nitrogen content with IAA treatment was also reversed by thiamine. B_1 was stimulatory in the initial stages and inhibitory at the later stages. IAA was inhibitory in the initial stages and stimulatory in later stages. This was a contrary behaviour indicating mutual

TABLE I

Oxygen uptake (*μl/seedlings*) *of the seedlings*
(The values are mean of two replications)

Treatment	Days after sowing 2	4	6	8
Control	6·0	40·0	25·5	18·0
B_1	15·0	43·0	49·0	19·0
IAA	11·0	26·0	18·0	12·0
B_1 + IAA	35·0	30·6	17·0	14·4

TABLE II

Nitrogen fractions (*mg/gm dry wt.*) *of the seedlings*
(The values are mean of three replications)

Treatment	Days after sowing 4 Total nitrogen	Protein nitrogen	Soluble nitrogen	6 Total nitrogen	Protein nitrogen	Souble nitrogen
Control	49·56 ±0·14	28·56 ±0·11	21·00 ±0·14	61·32 ±0·14	21·84 ±0·32	39·48 ±0·14
B_1	52·08 ±0·13	34·86 ±0·15	17·22 ±0·13	51·24 ±0·37	30·24 ±0·75	21·00 ±0·37
IAA	43·68 ±0·10	29·40 ±0·27	14·28 ±0·10	57·54 ±0·12	23·10 ±0·21	34·44 ±0·12
B_1 + IAA	53·34 ±0·30	29·82 ±0·13	23·52 ±0·30	57·96 ±0·17	23·94 ±0·87	34·02 ±0·17

antagonistic nature of both. B_1 antagonised the efficacy of IAA in increasing protein nitrogen through decreasing the soluble nitrogen. Soluble nitrogen content was not utilized in protein synthesis in IAA treated seedlings. That IAA was inhibitory in the initial stages and promotory at the later stages can be corroborated by the earlier report of Evans and Ray[1]. The role of thiamine in respiration in the conversion of pyruvate to acetyl CoA is well known. The possibility that B_1 may cause reduction in endogenous level of IAA cannot however be ruled out.

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Sri Venkateswara University, N. Raja Kumar.
Tirupati, *March* 11, 1976.

---

1. Evans, M. L. and Ray, P. M., *J. Gen. Physiol.*, 1969, 53, 1.
2. Dahlhelm, H., *Planta,* 1956, 86, 224.
3. Wain, R. L. and Fawcett, C. H., *Plant Physiology,* Ed. F. C. Steward, 1969, 5 A, 272.
4. Markham, R., *Biochem. J.*, 1942, 36, 790.
5. Thimann, K. V. and Loos, G. M., *Plant Physiol.,* 1957, 32, 274.

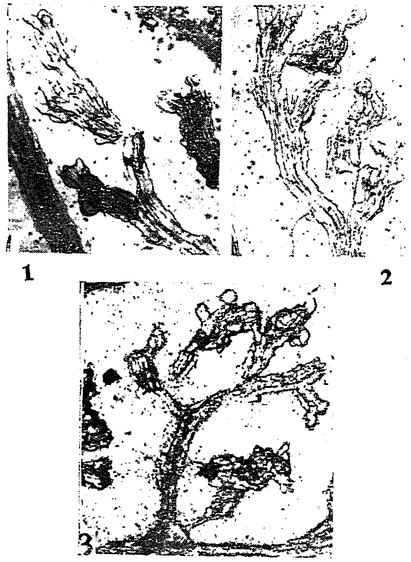
ON TERMINAL SCLEREIDS IN *BELLENDENA MONTANA* R.Br. (PROTEACEAE)

During the studies on the comparative morphology of foliar sclereids in angiosperms, a hitherto unreported case of a range in pattern of distribution of terminal foliar sclereids was observed in a remote member of the Proteaceae, which warranted a detailed study.

Materials

Bellendena montana R. Br. Tasmania, Summit of Mounts Wellington, Simson 688 (LE) ; Tasmania, Gunn. s.n. (LE) ; Nova Holland, Preiss 666 ((LE).

Cleared leaf sectors of the three specimens of *Bellendena montana,* a monotypic exclusively Tasmanian species have revealed very interesting association of sclereids at the vein endings despite the occurrence of thinly sclerosed sheathing cells along major and minor veins. The range in pattern of distribution of sclereids at the vein endings include (*a*) terminal spheroidal or vermiform sclereids in singles or twos and sometimes in threes in the submarginal periphery (Fig. 3), (*b*) a bunch of vermiform sclereids at the terminal or subterminal position in the proximal half of the leaf (Fig. 2), (*c*) vermiform sclereids overlapping or intertwined with each other in close juxtaposition with tracheids at the distal or subdistal portions of the forking minor veins and veinlets in the midhalf of the lamina (Figs. 1 and 2), (*d*) the same pattern as in (*c*) but showing 'feather brush' pattern in having free round ended sclereids aligned along the vein leaning towards the adjoining mesophyll cells in the vicinity of midrib region (Fig. 4). This diverse pattern in all probability is a mechanical reinforcement to reduce the detrimental effects of external pressures on the exposed laminar surface, and also to solve the problem of optimization of self-weight against rupture or curling and retention of rigidity and elasticity to regain its own shape once the force is withdrawn.



Figs. 1–3. Cleared laminae of *Bellendena montana* showing a range in pattern of distribution of terminal foliar sclereids, × 96.

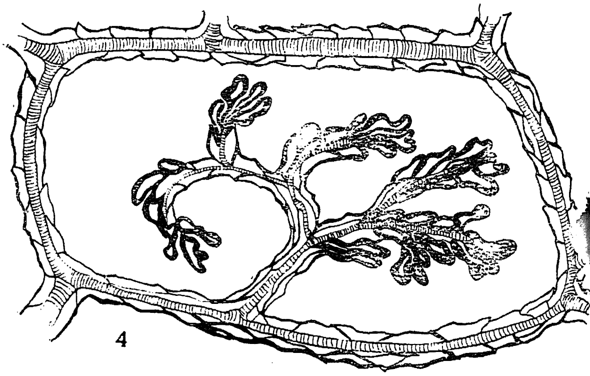


Fig. 4. Surface view of the lamina showing 'feather brush' pattern of foliar sclereids, × 60.

Transections show that sclereids in contact with the tracheary veins are mid-mesophyll in position, and often show irregular lobes of round endings and occasionally the lobes are proliferated. The cell wall is thick and exhibits pale yellow colour under light microscope. This depiction of colour is very characteristic in this species and the reason for it is as yet not known. Mature sclereids possess lumina of irregular width and are free from pits[8, 9] and contents.

Terminal sclereids of diverse pattern as described above is unique for this species. Their range of variation is a distinct diagnostic feature of this species. Up-to-date interesting topographic relationship with sclereids and vein-endings has been recorded in closely related and distinct taxa of angiosperms such as *Canella, Warburgia, Cinnamoma, Pleodendron* of the Canelliaceae[1], *Capparis, Niebuhria* of the Capparidaceae[2], *Salicornia* of the Chenopodiaceae[3, 4], *Pseudoconnarus* of the Connaraceae[5], *Cunonia, Weinmannia* of the Cunoniaceae[6], *Hibbertia* of the Dillenieaceae[7, 8], *Goupia* of the Goupiaceae[9], *Hamamelis, Exbucklandia, Eustigma, Rhodoleia, Disanthus, Liquidamber* of the Hamamelidaceae[2], *Cynometra, Cassia, Dimorphandra* of the Leguminoseae[10-12], *Aromodendron, Kmeria, Manglietia, Michelia* of the Magnoliaceae[2], *Coryphadenia, Memecylon, Mouriri* of the Melastomataceae[2], *Olinia* of the Oliniaceae[13], *Scherebera* of the Oleaceae[2], *Limomium* of the Plumbaginaceae[2], *Moutabea* of the Polygalaceae[2], *Bellendena* of the Proteaceae, *Bruguiera* of the Rhizophoraceae[2], *Scyphiphora* of the Rubiaceae[2], *Boronia* and *Boronella* of the Rutaceae[2], *Hannoa* of the Simarubaceae[2], *Enkleia, Linostoma* of the Thymelaeaceae[2], and *Vochysia* of the Vochysiaceae[14]. In all the abovementioned 41 taxa belonging to 22 families the terminal sclereids are distinct and clearly associated with veinendings despite their typological diversity. The 'terminal' position is confirmed by ontogenetic study only in 4 taxa, namely, *Mouriri, Memecylon, Boronia,* and *Niebuhria*[2] and in the rest this positional relationship is recognised purely on a topographical basis. Its occurrence in apparently near and widely reported taxa belonging to different orders, families, genera and species can be considered as a significant morphological aspect occurring independently at various levels in a phylogenetic classification. This unusual feature of the intimate relationship of tracheary endings with the thick walled idioblasts is intriguing and the exact physiological or mechanical advantage of this association to any particular environmental stress is as yet not proved.

We are grateful to the authorities of V. I. Kumorov, Botanical Institute, Leningrad, for the gift of leaf-specimens used in the present study. Furthermore, we express our thanks to Sri N. K. Das for elucidating the mechanical reinforcement advantages involved in the construction of leaves.

Botanical Survey of India, T. ANANDA RAO.
P.O. Botanic Garden, MRS. SILPI DAS.
Howrah 3, *March* 18, 1976.

1. Parmeswaran, N., *Proc. Indian Acad. Sci.*, 1961, **54 B**, 306.
2. Rao, T. A. and Bhupal, O. P., *Adv. Pl. Morph.*, 1972, p. 46.
3. Fahn, A. and Arzee, I., *Amer. J. Bot.*, 1959, **46**, 330.
4. De Frain., *J. Linn. Soc. Bot.*, 1912, **41**, 317.
5. Dickison, W. C., *Elisha Mitchell Sci. Soc.*, 1973, **89**, 121.
6. —, *Bot. J. Linn. Soc.*, 1975, **71**, 275.
7. —, *J. Arnold Arbor.*, 1969, **50**, 384.
8. —, *Ibid.*, 1970, **51**, 89.
9. Rao, T. A. and Bhattacharya, J., *Curr. Sci.*, 1975, **44**, 132.
10. —, *Ph.D. Thesis*, Mysore University, India, 1953.
11. Banerjee, M. L. and Sirkar, G., *Bull. Bot. Soc.*, Bengal, 1974, **28**, 1.
12. Ragonese, A. M., *Bot. J. Linn. Soc.*, 1973, **67**, 225.
13. Mujica, M. B. and Cutler, D. F., *Kew Bull.*, 1974, **29**, 93.
14. Rao, T. A. and Mody, K. J., *Proc. Indian Acad. Sci.*, 1961, **53 B**, 257.

REVERSAL OF NIACIN INHIBITED GROWTH BY GIBBERELLIC ACID IN GREEN GRAM (*PHASEOLUS RADIATUS* L.)

THE possibility of the regulation of plant growth by natural or synthetic antagonists of gibberellic acid (GA) remains only an interesting possibility to date. A synthetic diterpene, chlorofluorenol was reported by Ziegler *et al.*[1] as a possible GA antagonist but Krelle[2] negatived this proposition. Several unidentified natural inhibitors have been suggested as GA antagonists[3-6]. The ability of some synthetic growth retardants to inhibit GA synthesis is not to be construed as a gibberellin antagonism, even though their inhibition may be relieved by added GA[7]. The present study has been designed to know the action of niacin (*a vitamin of the B group*) on gibberellic acid in relation to growth.

Seeds of green gram (variety 525) were subjected to soaking in 50 ppm GA and 1000 ppm niacin for 24 h. in dark in petri dishes before sowing. After this interval the seeds were allowed to germinate in distilled water for about 8 days. Growth and respiratory activity was measured by the usual techniques. Nitrogen fractions were estimated according to the methods of Markham[8], and Thimann and Loos[9].

Niacin (1000 ppm) inhibited the growth by 60–80% (Fig. 1). It is to be noted in this connection that other B group vitamins are not inhibitory at this concentration but instead stimulatory, at least initially. GA (50 ppm) stimulated the growth to about 40 to 50% over the control. It is quite interesting to note that when GA (50 ppm interacted with niacin (1000 ppm), the inhibitory effect of niacin was nullified and there was about 900% stimulation of growth. This apparently indicated niacin is blocking the synthesis of endogenous GA. When GA was added exogenously, the GA synthesis must have been restored. Arditti[10] assumed that the presence of niacin may promote NAD and NADP biosynthesis which in turn may stimulate the production and utilization of a variety of compounds. In the present study it is quite possible that niacin has stimulated the gibberellin activity, resulting in 900% enhancement of growth over the control. The growth-promoting effect of niacin may, therefore, be a rather indirect one, the improved growth being only an observable index of a considerable number of biochemical—physiological—events in the seedlings. Niacin must have been activated by GA. This was corroborated by the reversal of respiratory inhibition with niacin by GA (Table I). Protein nitrogen content was higher in niacin treated seedlings. Combination of GA with niacin enhanced the protein nitrogen content much more than that of control, niacin and GA treatments (Table II).

TABLE I

O_2 uptake (μl/seedling)

(Mean of two replications)

Treatment	Days after sowing 2 days	4 days	6 days	8 days
Control	6·0	43·0	25·5	18·0
Niacin	12·20	17·0	9·72	16·76
GA	33·0	35·0	24·0	25·0
GA + Niacin	15·0	33·0	34·0	16·0

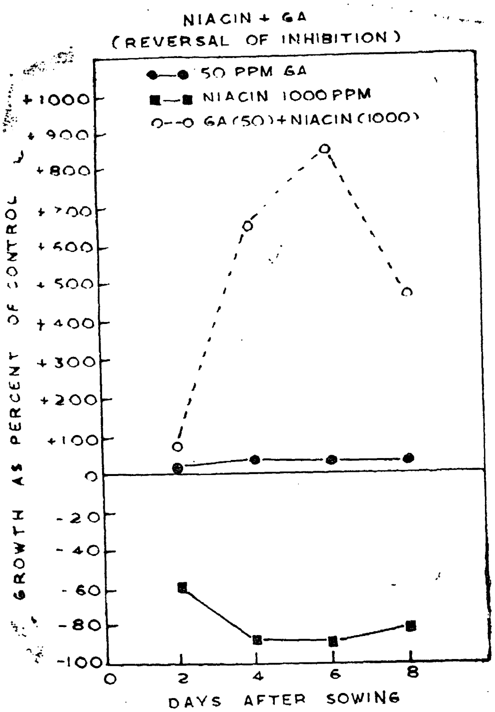


FIG. 1

TABLE II

Nitrogen fractions (mg/g dry wt) of the seedlings

(The values are the mean of three replications)

Treatment	4 Days Total Nitrogen	 Protein Nitrogen	 Soluble Nitrogen	6 Days Total Nitrogen	 Protein Nitrogen	 Soluble Nitrogen
Control	49·560 (0·792)	28·560 (0·456)	21·000 (0·336)	61·320 (0·861)	21·840 (0·307)	39·480 (0·554)
GA (50 ppm)	53·760 (0·933)	29·820 (0·518)	23·940 (0·415)	56·280 (0·768)	22·680 (0·309)	33·600 (0·459)
Niacin (1000 ppm)	44·520 (0·847)	30·240 (0·576)	13·280 (0·271)	48·720 (0·826)	25·200 (0·428)	23·520 (0·398)
GA + Niacin (50 + 1000 ppm)	46·620 (0·763)	33·600 (0·550)	13·020 (0·213)	50·400 (0·669)	29·820 (0·396)	20·580 (0·273)

The results in the parenthesis indicate mg/organ, This apparently indicates that GA activates the effect of niacin. There are reports to show that these natural growth substances, *viz*, gibberellins, IAA and cytokinins activate vitamin activity. Digby and Skoog[11] showed cytokinin activation of thiamine biosynthesis in tobacco callus cultures. Thus, the final enhancement of growth to 900% with niacin and GA interaction appears to be synergistic although niacin is antagonistic to the synthesis of gibberellin endogenously. That the endogenous level of GA is inhibited is evident from growth inhibition by niacin and its reversal by added GA. Finally, it is concluded that niacin at higher concentrations (unlike other vitamins) can inactivate GA thus inhibiting the growth.

The authors thank Prof. V. S. Rama Das for his encouragement and providing the facilities.

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Sri Venkateswara University, N. RAJA KUMAR.
Tirupati, *March* 22, 1976.

---

1. Ziegler and Luttage, V., *Planta*, 1966.
2. Krelle, E., *Plant Growth and Development* (Ed. A. C. Leopald, pp. 146) 1967, 70, 193.
3. Corcoran, M. R., West, C. A. and Phinney, B. O., *Adv. Chem.*, 1961, **28**, 152.
4. Kohler, D., *Planta*, 1964, **63**, 326.
5. Michniewics, M. and Kopcewicz, J., *Rosszniki Nauk Rolu*, 1966, **90**, 689.
6. Thomas, T. H., Wareing, P. F. and Robinson, P. M., *Nature*, 1965, p, 205.
7. Cathey, H. M., *Ann. Rev. Plant Physiol.*, 1964, **15**, 271.
8. Markham, R., *Biochem. J.*, 1942, **36**, 790.
9. Thimann, K. V. and Loos., G. M., *Plant Physiol.*, 1957, **32**, 274.
10. Arditti, Joseph, *Amer. Jour. Bot.*, 1967, **54**, 291.
11. Digby, J. and Skoog, F., *Plant Physiology*, 1966, 41, 647.

LEPTURUS RADICANS (STEUD.) A. CAMUS—A NEW RECORD FOR INDIA

DURING studies on Poaceae of Karnataka State the authors came across 4 herbarium specimens of *Lepturus repens* (G. Forst.) R. Br., all belonging to a single collection of R. K. Bhide from Karka Forest, Naka, near Halyal in North Kanara District of Karnataka State dated 9th February, 1920 and the same are cited by Blatter and McCann (1935)[1]. But on a scrutiny it was found that these specimens belong to another species, *viz. L. radicans* (Steud.) A. Camus. A consultation of *BSI* herbarium and literature revealed that this species is not reported from India so far and hence the present note. The above-stated two species differ from each other in the following characters:

	L. radicans (Steud.) A. Camus	*L. repens* (G. Forst.) R. Br.
1.	Spikes 3–5 cm long	Spikes 5–15 cm long
2.	Spikelets 3–5 mm or the terminal 5–8 mm long	Spikelets 5–14 mm or the terminal upto 18 mm long
3.	Upper glume ovate-oblong or lanceolate-oblong, acute or acuminate but not drawn out into a short arista.	Upper glume lanceolate, tailed or finely acuminate
4.	Lemma 2·5–4 mm long	Lemma 4–5·5 mm long

Bor (1960)[2] gives its distribution as Madagascar, Mascarene Islands, Tanganyika Territory and Ceylon, which, with the present report, now extends to India as well. A brief description has been given by Senaratna (1956)[4] and Clayton *et al.* (1974)[3]. The former has given the illustrations as well. However, the description given by Blatter & McCann (*l.c.*)[1] under *L. repens* (G. Forst.) R. Br. belongs to that species only but his citation of the above-said Bhide's specimens now belongs to *L. radicans* (Steud.) A. Camus.

The authors are thankful to Dr. R. S. Raghavan, Regional Botanist, Botanical Survey of India, Western Circle, Poona, for facilities and to Dr. K. Subramanyam, Ex-Director, Botanical Survey of India, for encouragement.

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---

1. Blatter, R. and McCann, C., *The Bombay Grasses*, Manager of Publications, Delhi, 1935, p. 279.
2. Bor, N. L., *The Grasses of Burma, Ceylon, India and Pakistan*, Pergamon Press, Oxford, London, New York, Paris, 1960, p. 585.
3. Clayton, W. D., Phillips, S. M. and Renvoize, S. A., *The Flora of Tropical East Africa—Gramineae* (Part 2), Whitefriars Press Ltd., London and Tonbridge, 1974, p. 389.
4. Senaratna, S. D. J. E., *The Grasses of Ceylon*, Government Press, Ceylon, 1956, p. 62.

MALE STERILITY IN SUNNHEMP (*CROTALARIA JUNCEA* L.)

MALE sterility in *Crotalaria striata* has been reported[1,2] in literature. The present note deals with the male sterility in sunnhemp.

Meiosis and the subsequent tetrad formation in the case of the sterile sunnhemp plant were found normal but the pollen was contorted and devoid of any nuclear content (Fig. 1). Periodic examinations also revealed the consistency of the degenerating pollen.

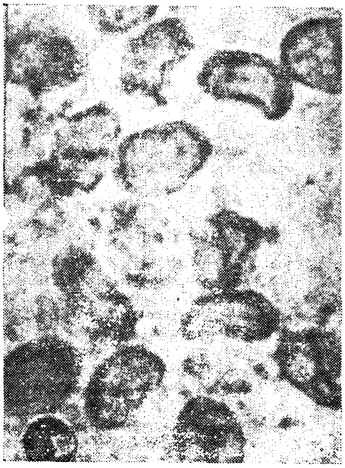

FIG. 1

This plant was allowed to set seeds openly and the progeny gave both male fertile and sterile plants. The species being predominantly a cross-fertilizer[3,4] the occurrence of two such types in the hybrid generation tentatively indicates that the male sterlity may either be of genie or gene cytoplasmic type. Further study which is under progress may give a clear picture of its inheritance.

The author is highly thankful to Dr. S. L. Basak, Head of Division, Plant Breeding and Genetics, J.A.R.I., for helpful suggestion.

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---

1. Kempanna, C. and Sastry, K. S. K., *Curr. Sci.*, 1958, **27**, 181.
2. Edwardson, J. R., *J. Hered.*, 1967, **58**, 266.
3. Mitra, G. C., *Broteria*, 1970, **39**, 1–2, 79.
4. Mohan, K. V. J., *Ind. J. Genet. and Plant Breeding*, 1971, p. 31.

A NEW HOST RECORD FOR *BRANCHIOMYCES SANGUINIS* PLEHN.

DURING the course of an investigation on fungi associated with fish diseases, the adult individuals of *Channa marulius* (Bl.) were found to be suffering from branchiomycosis during April–May, 1976, at Government Fish–Farm, Gujar Tal, Jaunpur. The fishes were observed gasping for air on the surface of water for certain period of time after which they usually died. Observations on gills of these fishes revealed the destruction and discoloration of gill lamellae (Fig. 1).

The pathogen was identified as *Branchiomyces sanguinis* Plehn, a phycomycete having 10·5–28·5 μ wide hyphae with spores 6–10·8 μ in diameter.

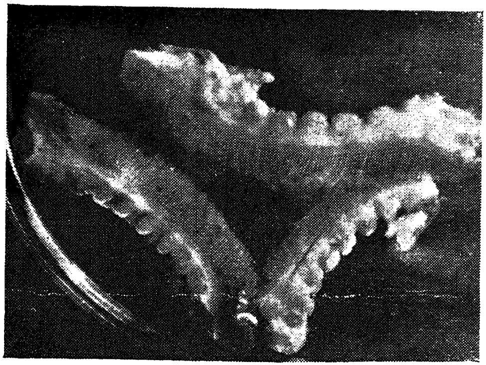

FIG. 1. Branchimycosis (gill rot) of the gills of *Channa marulius*, (Note the eroded ares).

A perusal of the literature[1,2] has revealed that the present communication is the first report on branchiomycosis (gill rot) in *Channa marulius*, which is an important edible fish of eastern U.P.

We are indebted to the authorities of C.S.I.R. for financial assistance and to Dr. Y. B. Singh, Principal, St. Andrew's College, Gorakhpur, for providing facilities for the work.

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---

1. Reichenbach Klinke, H., *Fish Pathology*, T.F.H. Publ., Inc., Ltd., Neptune, 1973.
2. Khan, H. A. and Jhingran, V. G., *Synopsis of Biological Data on Rohu* [*Labeo rohita* (*Hamilton, 1822*)], FAO Fisheries Synopsis No. 11, 1975.

A NOTE ON THE NATURAL OCCURRENCE OF *CERCOSPORA ORYZAE* MIYAKE IN RICE VARIETIES IN TAMIL NADU STATE

AMONG the many diseases of rice crop, the narrow brown leaf spot caused by *Cercospora oryzae* Miyake was observed as early as in 1906 by Metcalf in USA and described by Miyake in 1910 from Japan[1]. Considered as a disease of minor importance, it was first noticed around Annamalainagar area in Tamil Nadu in 1960 on '*indica*' rice varieties[2]. The disease incidence increased markedly after 1965, with the spread of several high yielding rice varieties of '*japonica*' × '*indica*' types. Visits to important rice growing areas in the State in different seasons during the last three years revealed the widespread prevalence of the disease on most of the popular varieties and heavy lesions were noticed even during the summer period. The diseased specimens were collected, examined and presence of the fungus was confirmed. On the basis of the length of the lesions and their number per sq. cm leaf area, a disease-index was prepared and on the basis of the index the rating of the rice varieties is as follows:

Resistant	Moderately resistant	Moderately susceptible	Susceptible	Highly susceptible
Mashuri (0)	TKM–6 (5)	Jaya (15)	Karuna (45)	ADT–31 (75)
Bhavani (0)	Pusa–33 (7)	Vani (15)	Vaigai (46)	Kanchi (75)
GEB–24 (0)		Bala (15)		
	IR-22 (7)		T(N)–1 (47)	IR-20 (80)
IR–26 (0)		IR–8 (16)		Kannagi (80)
IR–28 (0)	Co. 31 (7)	IR-24 (20)	Co.25 (48)	ADT.–6 (80)
IR–29 (0)	Sona (8)		BCP–1 (49)	
IR–30 (0)	PTB. 10 (8)	SLO-16 (23)		
		Co. 39 (23)		
Co. 32 (2)		ASD. 7 (23)		
Co. 36 (2)				
Basumathi (2)		PTB.-15 (25)		
Kattisamba (2)				

Data within parenthesis indicate percentage of incidence.

While varieties like Mashuri and Bhavani are resistant to the disease, others like IR-20 and ADT-31 are highly susceptible. Most of the wild species of rice are free from this disease and probably this is the reason for the resistant nature in the latest rice varieties from the International Rice Research Institute, Los Banos, the Philippines, *viz.*, IR-28, IR-29 and IR-30, since the genetic characters of *Oryza navira* are carried by them.

A study of the mechanism of the disease resistance, in rice varieties is in progress.

The author is highly indebted to Dr. G. Rangaswami, Vice-Chancellor, Tamil Nadu Agricultural University, Coimbatore-3, for his generous help and guidance in the preparation of this paper.

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---

1. Ou, S. H., *Rice Diseases*, CMI, Kew, U.K., 1972. p. 213.
2. Rangaswami, G., Seshadri, V. S. and Channamma, K. A. L., *Fungi of South India*, Univ. of Agri. Sciences, Bangalore, 1970, p. 20.

REVIEWS AND NOTICES OF BOOKS

Tbe Physiology of Flowering Plants. (Second Edition). By H. E. Street and H. Opik. (Edward Arnold, 25, Hill Street, London W1 XB LL). Pp. viii + 280. Price : £ 9.00.

Tbe Physiology of Flowering Plants : *Their Growth and Development,* II Edn. By H. E. Street and Helgi Opik. Published in series of student texts in Contemporary Biology. General Editors : Professor E. J. W. Barrington, F.R.S. and Professor Arthur J. Willis. (Edwin Arnold Publishers Ltd., 25, Hill Street, London), 1976. Pp. 280. Price : £ 4.50.

The book attempts to provide a modern and comprehensive treatment of the developmental aspects of plant physiology. Written by two competent men in the field the book appears to have been very well received by both students and researchers in plant physiology.

When compared to the original 1970 text, the Chapters 1, 4, 10, 11 and 12 (replacing the original Chapter 11) in the book have been appropriately revised and updated. Out of the twelve chapters in 280 pages, only a few pages are devoted to aspects of cell structure and cell physiology. If these aspects were to have been dealt with in some more detail, so as to give the needed essentials for an appropriate consideration of the focal theme of the book, namely the major aspects of growth and development, the attempt would have been more meaningful. In fact some chapters in the book (Chapters 2, 8, 9, 10, 11 and 12) are devoted for aspects of growth and development. All these are very well written. They not only deal with growth and development of plant and its organs but also give detailed information on processes of cell division, cell enlargement, cell differentiation, cellular processes involved in the initiation of organs, organ growth and overall development of the plant body.

The chapter on germination is a master account. It introduces a number of fundamental processes which are treated in detail subsequently. The background thus established forms the basis for the later chapters.

Although each chapter in the book is linked to the other relevant ones by cross-references repetitions are limited. The subject-matter is treated with great competence and an attempt is made to render each chapter a self-contained unit. The selected references at the end of each chapter are most welcome and they no doubt trigger the student for further study. It is very true as the authors themselves state that "when a large body of knowledge is surveyed in a book of limited size, shortcomings are inevitable". For advanced students in the field, it should be said that the book gives only the primary data intended to provide an essential background. This is especially so with regard to tips on mineral nutrition, vernalization, fertilization, embryo development and seed dormancy.

On the whole one should accept that the book certainly gives a good background of developmental aspects of plant physiology to students and teachers concerned with the courses on Physiology of Flowering Plants : their growth and development.

D. A. GOVINDAPPA.

Irreversible Thermodynamics and the Origin of Life (*Proceedings of the Third International Biophysics Conference, MIT*). (Gordon and Breach, New York), 1975. Pp. xi + 69. Price : £ 3.90.

This conference report is a collection of diverse research papers presented at a Conference. The papers are interesting in themselves but become more valuable in the biophysical context. The papers of Ahron Katchalsky and of Ilya Prigogine deal with the behaviour of coupled diffusion and chemical reaction processes, Prigogine makes an eloquent presentation of symmetric breaking in a non-linearly coupled system leading to self-organization. Supplementary to this is the paper of Harold Morowitz who demonstrates a number of general results on the functioning of a non-equilibrium system. H. H. Pattee raises a number of tantalizing questions in his paper but his reasoning has mostly escaped this reviewer. The most valuable part of the proceedings is an informal discussion session with about a dozen participants where a number of interesting questions were discussed : like prebiotic evolution and whether laboratory experiments under atypical conditions had anything to do with it ; the potential for life in the solar system and the relevance of Mars probes; whether biology is a reductionist Science ; Geochemical evidence on early stages of life, the role of non-equilibrium thermodynamics to life processes.

E. C. G. SUDARSHAN.

Climatological Aspects of the Composition and Pollution of the Atmosphere. By G. C. Holzworth. (Technical Note No. 139. World Meteorological Organization, Geneva), 1974. Pp. 43. 17 figures.

The Technical Note is the outcome of a survey on climatological procedures for estimating the air-pollution potential of locations or areas, by the author in his capacity as COSAMC (WMO Commission for Special Applications of Meteorology and Climatology) *Rapporteur* on Climatologiical Aspects of the Composition and Pollution of the Atmosphere. The report offers some practical guidelines on the processing and use of regular surface and upper-air observations in terms of their climatological influence on the transport and diffusion of air pollutants.

The "climatological potential" for air pollution is concerned with what would happen to pollutants while they are airborne, specially to their concentration, if they were emitted to the atmosphere. It impiles that climatological data can be evaluated in advance of actual pollutant emissions, in order to foretell the meteorological consequences of such emissions. The important processes that occur in the atmosphere include transport, diffusion, precipitation scavenging, gravitational fallout, impaction, pollution transformation (including those induced by solar radiation), etc. The meteorological variables of importance are windspeed and direction, static stability, mixing heights, diffusion parameters, fog and precipitation. A more general variable is atmospheric stagnation which refers to meteorological conditions that are often associated with or conducive to episodes with relatively high concentrations of pollutants, especially as experienced in cities. Various definitions and applications of stagnation are described for different parts of the world. Unfortunately the data are limited to temperate and northern latitudes of the northern hemisphere due to a paucity of available meteorological studies relating to air pollution in the tropics. This is most unfortunate and deserves urgent correction, particularly since these are the areas of the world where industrialization, energy consumption and pollutant emission are likely to increase most rapidly.

The most compelling reason to develop an air-pollution potential climatology for a city region or country is to provide guidelines for the efficient management of pollutant emissions from both existing and new sources. Climatology can be used in the development of landuse ordinances, the location of motor vehicle thoroughfares, airports, ports and large single sources of pollution. Although, thorough evaluation of the siting of large specific sources of pollution often requires detailed information obtained at the site, climatological information for nearby locations can be very useful in preliminary planning.

In addition to transport and diffusion and air quality, some attention is also devoted to climatological influences on the transformation of pollutants while they are airborne, especially the photochemical formation of oxidant. Nearly 100 references and a brief summary of pertinent publications of the WMO adds to the value of the report.

Utilisation of Aircraft Meteorological Reports. By S. Simplicio. (WMO No. 400, Technical Note No. 141, Secretariat of the World Meteorological Organization, Switzerland), 1975. Pp. 34, 9 figures.

Meteorological observations made aboard aircraft in flight are of great value to meteorologists in supplementing synoptic weather data available from other sources. This Technical Note is a revised edition of two earlier Technical Notes prepared in 1964 and 1966, on a directive from the Executive Committee of the WMO. The original note was perpared by Working Groups of the Commission for Aeronautical Meteorology of which Mr. Simplicio was a member and later Chairman. The present note is prepared entirely by Mr. Simplicio.

The Technical Note supplies the reader with information regarding the various meteorological uses to which the observations made aboard aircraft may be put and describes the techniques to be used in order to derive maximum benefit from the information. The Note begins with a short review of the current observing and reporting procedures for aircraft observations and an evaluation of the elements included in AIREPs or Air reports. These present weather, frontal passage, D-value, turbulence and icing, bases and of clouds and weather radar echoes.

Chapter 2 deals with the use of AIREPs in numerical weather analysis and prediction. The use of temperature, wind, turbulence, icing, pressure, weather and cloud data obtained from all reports in analysing upper air charts is described, with model upper air charts prepared following the method described.

Chapter 3 describes the particular use of AIREPs in providing assistance to air navigation, in preflight planning documentation and briefing, en-route forecasts and short term forecasts of terminal weather condition.

The last chapter deals with other uses of air reports, *i.e.*, statistical analysis for climatological summaries, technical investigations and verification of forecasts. An Appendix describes Corrections of D-values for non-standard temperature conditions. A bibliography completes the report.

The Technical Note is a very useful, though rather elementary account of a type of weather observation, not extensively used at present. It is particularly valuable in areas where the network of surface and upper air observations is sparse, as over the oceans, and, often provides the only direct information of meteorological developments over the area. It is the only source of information on the actual occurrence of turbulence and icing. It is hoped that this method will come into more widespread use in the near future particularly during the FGGE and Monex.

INDIAN NATIONAL SCIENCE ACADEMY, NEW DELHI

At the Annual General Meeting of the Indian National Science Academy held on Friday, the 8th October, 1976, Dr. R. Ramanna, Director, Bhabha Atomic Research Centre and Professor, Tata Institute of Fundamental Research, Bombay, was elected President of the Academy, for the year 1977.

The other officers elected for the ensuing year were: Vice-Presidents: Prof. R. C. Mehrotra, University of Delhi, and Prof. A. K. Sharma, University of Calcutta; Treasurer: Dr. M. L. Dhar, Ex-Director, Central Drug Research Institute, New Delhi; Foreign Secretary: Dr. A. R. Verma, National Physical Laboratory, New Delhi; Secretary: Dr. M. G. Deo, AIIMSc., New Delhi.

The other officers re-elected for the year 1977 were: Secretary: Prof. A. N. Mitra, University of Delhi; Editors of Publications: Prof. L. S. Kothari and Prof. K. N. Saxena.

The members of Council elected (or re-elected marked *) were: Prof. B. K. Bachhawat,* IIEM, Calcutta; Dr. V. G. Bhide,* NPL, New Delhi, Prof. A. Bose,* IACSc., Jadavpur, Calcutta; Prof. B. G. Deshpande, University of Poona; Prof. P. C. Dutta,* IACSc., Jadavpur, Calcutta; Prof. J. Ganguly, Indian Institute of Science, Bangalore; Prof. M. S. Narasimhan, TIFR, Bombay; Dr. Y. Nayudamma, CSIR, New Delhi; Dr. N. Parthasarathy,* Madras; Prof. V. Puri, Meerut University; Dr. G. S. Ramaswamy, CSIR Campus, Madras; Prof. C. N. R. Rao,* IIT, Kanpur; Prof. S. N. Sarkar,* Dhanbad; Prof. G. P. Sharma, Punjab University; Dr. A. Sreenivasan,* Bombay; Dr. K. Sundaram,* BARC, Bombay; Prof. S. K. Trehan, Punjab University; Dr. H. V. K. Udupa,* CECRI, Karaikudi.

Fellows elected at the Annual General Meeting: Prof. T. N. Ananthakrishnan, Loyala College, Madras; Prof. (Mrs.) Archana Sharma, University of Calcutta; Dr. R. D. Asana, Emeritus Scientist, Valsad; Prof. D. P. Burma, Banaras Hindu University; Prof. G. B. Deodikar, MACS, Poona; Dr. L. K. Doraiswamy, NCL, Poona; Prof. T. K. Ghosh, Calcutta; Prof. M. K. Jain, IIT, New Delhi; Dr. G. C. Jain, NPL, New Delhi; Shri L. N. Kailasam, GSI, Calcutta; Dr. S. V. Kessar, Panjab University; Dr. T. N. Khoshoo, NBG, Lucknow; Dr. (Mrs.) Usha K. Luthra, ICMR, New Delhi; Miss Anna Mani, Meteorological Office, New Delhi; Dr. Sushil Kumar Mukherjee, Bose Inst[illegible] Calcutta; Dr. K. Nagarajan, CIBA, Bombay; Prof. K. M. Naha, IIT, Kharagpur, Prof. J. V. Narlikar, TIFR, Bombay; Dr. H. L. Nigam, University of Allahabad, Prof. B. R. Puri, Punjab University; Dr. S. Z. Qasim, NIO, Goa; Prof. L. K. Ramachandran, Osmania University, Hyderabad; Prof. S. Ramanan, TIFR, Bombay; Dr. (Mrs.) Kamal J. Ranadive, Tata Memorial Centre, Bombay; Prof. O. Siddiqi, TIFR, Bombay; Prof. P. N. Srivastava, JNH, New Delhi; Prof. B. V. Thosar, TIFR, Bombay; Dr. L. R. Vaidyanath, Ind. Copper Information Centre, Calcutta; Prof. G. Venkataraman, Reactor Research Centre, Kalpakkam; Dr. Yash Pal, Ahmedabad.

Foreign Fellows Elected:

Prof. Viktor Amazaspovich Ambartsomian, (USSR), Prof. S. N. Carey, Prof. S. Hammond George, gard Paul (C. openhagen, Denmark), Dr. R. Odhiambo gard Paul (Copenhagen, Denmark), Dr. R, Obhaiambo (Thomas, Nairobi, Kenya), Prof. Laurent Schwarte, (Paris 5C, France), Prof. Uvais Siddeek Sultanbawa Mohammed, Univ. of Sri Lanka.

'Science Academy Medals for Young Scientists' —Awardees for 1976

Dr. Srikumar Banerjee, BARC, Bombay; Dr. (Mrs.) Julie Banerji, University College of Science, Calcutta; Dr. A. R. Bhattacharya, Lucknow University; Dr. S. G. Dani, TIFR, Bombay; Dr. Shamim Haider, Banaras Hindu University; Dr. R. V. Halyburton, Oil India Ltd., Assam; Dr. (Miss) Priti Mohapatra, Utkal University, Bhubaneswar; Dr. V. M. Pawar, Marathwada Agricultural University; Dr. S. S. S. V. Prasad, The University of Texas, Texas; Dr. Abhijit Sen, PRL, Ahmedabad; Shri A. Surolia, Medical College, Vellore; Dr. V. V. S. Tyagi, Panjab Agricultural University, Ludhiana.

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